# TECHNICAL MEMORANDUM gdc

## BIOASSAY TESTING – JOINT CANNERY OUTFALL EFFLUENT FEBRUARY 2007 SAMPLING

**Prepared For:** StarKist Samoa (NPDES Permit AS0000019)
COS Samoa Packing (NPDES Permit AS0000027)

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**Date:** 27 July 2007

**Distribution:** Carl Goldstein
United States Environmental Protection Agency, Region 9
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American Samoa Environmental Protection Agency

### SUMMARY

The NPDES permits for StarKist Samoa and COS Samoa Packing require semi-annual whole effluent toxicity testing. The test was conducted for the 2007 non-tradewind season using mysid shrimp and effluent samples collected February 27th and 28th, 2007. Results indicated an $LC_{50}$ of 44.1% effluent. This is within the range of previous tests. The result represents effluent toxicity of 2.7 TUa (acute toxicity units). Based on initial dilution modeling for critical conditions[1], the toxicity will be reduced to non-toxic levels (0.3 TUa) within 2.2 meters of the discharge, within 2 seconds, and less than 0.5 m above the diffuser or approximately 53 m below the water surface. This is well within the zone of initial dilution (ZID) for critical conditions. At the edge of the ZID the toxicity is reduced to less than 0.01 TUa.

[1] See "Request for Water Quality Certification and the Definition of Mixing Zones". gdc, 28 June 2007

## INTRODUCTION

This memorandum presents the results of the supplementary bioassay testing of the Joint Cannery Outfall effluent sample that was collected in February 2007[2]. The testing is required by the NPDES Permits that became effective in January 2001. The February 2007 test is the thirtieth test conducted since toxicity testing of the Joint Cannery Outfall effluent began in 1993[3].

### Study Objectives

Section D.1 of the StarKist Samoa and COS Samoa Packing NPDES Permits requires that semiannual definitive acute bioassays (96-hour static bioassays) be conducted on the cannery effluent. The purpose of these tests is to determine whether, and at what effluent concentration, acute toxicity may be detected for the combined joint cannery effluent discharge into Pago Pago Harbor.

### Study Approach

The U.S. Environmental Protection Agency (USEPA) has conducted a number of reviews of the effluent sampling, analysis, and bioassay tests conducted in the past. All comments from USEPA have been incorporated into the sampling and sample handling standard operating procedures (SOP) or have been incorporated into the procedures used by the laboratory doing the test. The comments, responses, and SOP have been documented in previous reports.

The NPDES permit conditions require that the bioassay tests be conducted with the white shrimp, *Penaeus vannami* (postlarvae). In the event *Penaeus vannami* is not available at the time of the tests, the permit specifies the substitute species, *Mysidopsis bahia*, which now has been renamed *Americamysis bahia*. For the February 2007 samplings, *Penaeus vannami* was not available and *Americamysis bahia* was used.

Effluent samples were collected from the StarKist Samoa and COS Samoa Packing facilities at three hour intervals over a 24-hour period. The acute effluent bioassay test was conducted using a combined, flow-weighted, composite effluent sample made up from the effluent samples from both canneries, as allowed by the NPDES permit conditions. This combined effluent bioassay is representative of the wastewater discharged from the joint cannery outfall to Pago Pago Harbor.

[2] The semi-annual joint cannery outfall effluent bioassay tests are performed during the Non-Tradewind and Tradewind oceanographic seasons.

[3] Testing was not conducted during 1999. Extra tests using two organisms were conducted in March 1995 and February 1996. A supplementary test was conducted in May 2006.

## EFFLUENT SAMPLING METHODS

The February 2007 effluent samples were collected between 09:00 on 27 February 2007 and 06:00 on 28 February 2007. A flow-weighted composite sample of final effluent was created from both the StarKist Samoa and COS Samoa Packing effluent discharges. Samples were collected from the established effluent sampling sites. Detailed sampling procedures are described in the SOP for cannery effluent sampling.

A total of eight grab samples were collected into 1-gallon plastic cubitainers at each cannery. Samples were collected at approximately three-hour intervals over the 24-hour period. The samples were stored on ice or in a refrigerator until the completion of the 24-hour sampling period. After all samples were collected a 5-gallon flow-proportioned composite sample was prepared. The grab sample collection times, effluent flow rates, and the relative effluent flow volumes calculated from plant flow records are summarized in Table 1. The relative effluent flow volumes were used to prepare the final composite sample, which was used to fill the sample container shipped to the laboratory for testing.

A 5-gallon cubitainer containing the composite sample was packed on ice in an ice chest for shipment to the laboratory. A chain-of-custody form for the sample was completed and sealed into a zip-lock bag and taped inside the lid of the ice chest. The sample was shipped via DHL to the testing laboratory. The chain-of-custody form and the DHL waybill for the test are provided in Attachment I.

**Table 1**
**StarKist Samoa and COS Samoa Packing**
**24-hour Composite Effluent Sample for Bioassay Testing**
**February 2007 Sample**

| Grab Sample Number | COS Samoa Packing | | StarKist Samoa | | Percent of Total Flow | |
|---|---|---|---|---|---|---|
| | Sampling Time | Effluent Flow (mgd) | Sampling Time | Effluent Flow (mgd) | Samoa Packing | StarKist Samoa |
| 27 February 2007 | | | | | | |
| 1 | 09:00 | 0.88 | 09:00 | 2.20 | 3.73 | 9.33 |
| 2 | 12:00 | 0.80 | 12:00 | 2.02 | 3.39 | 8.57 |
| 3 | 15:00 | 0.76 | 15:00 | 2.06 | 3.22 | 8.74 |
| 4 | 18:00 | 0.76 | 18:00 | 2.50 | 3.22 | 10.60 |
| 5 | 21:00 | 0.76 | 21:00 | 2.48 | 3.22 | 10.56 |
| 28 February 20077.46 | | | | | | |
| 6 | 00:00 | 0.76 | 00:00 | 1.76 | 3.22 | 7.46 |
| 7 | 03:00 | 0.76 | 03:00 | 2.16 | 3.22 | 9.16 |
| 8 | 06:00 | 0.76 | 06:00 | 2.15 | 3.22 | 9.12 |
| **Total** | | 6.24 [A] | | 17.34 [A] | 26.5% | 73.5% |
| **Mean** | | 0.78 | | 2.17 | Total = 100% | |

[A] Numerical total of column for calculation purposes. Total flow over a 24-hour period will be approximately the calculated mean.

## BIOASSAY TESTING PROCEDURES

EnviroSystems, Inc. located in Hampton, New Hampshire conducted the bioassay tests. The testing procedures and results of the bioassay tests are provided in the laboratory report included as Attachment II. This report summarizes the 96-hour acute bioassay tests conducted with reference to the USEPA document Methods for Measuring the Acute Toxicity of Effluents to Freshwater and Marine Organisms (EPA-821-R-02-012), 2002 as the source of methods for conducting the test. The bioassay tests were conducted considering and including USEPA's comments on previous bioassay tests, as documented in previous reports.

The test organisms were ≤ 5 days old and the test temperature was to be held at a nominal 20 °C. The actual temperatures ranged between 19°C and 20°C. Salinity was adjusted to 25 ppt at the start of the test and ranged between 25 and 28 ppt.

Demonstrated potential for a lethal immediate dissolved oxygen demand (IDOD) and a delayed dissolved oxygen demand spike (DDOD) had been discussed and documented in previous technical memoranda, which describe the first two tests conducted in 1993. Therefore, following an EPA approved modified testing protocol; all of the bioassay test chambers should be continuously aerated during the bioassay tests to maintain adequate levels of dissolved oxygen (DO)[4]. The test should also be renewed with pre-oxygenated effluent sample at 48 hours.

The DO levels were between 4.6 mg/l and 7.4 mg/l for the initial portion of test and between 5.9 mg/l and 7.3 mg/l following renewal. The DO levels were directly related to the percent effluent in the test chambers. Examination of the data indicates that the test results did not appear to be significantly influenced by DO concentrations.

Bioassay tests were carried out for effluent concentrations of 100, 75, 50, 25, 12.5, and 6.25 percent in seawater. Water quality was monitored daily and parameters measured included DO, pH, salinity, and temperature. Total residual chlorine and ammonia were also measured. Water quality data are provided in the Laboratory Report (Attachment II).

Reference toxicant tests using sodium dodecyl sulfonate (SDS) are conducted regularly by ESI with the relevant tests completed on 20 February 2007 for which the results were within the acceptable range based on the 20 most recent laboratory reference toxicant tests.

[4] The high initial dilution of the actual effluent discharge (>100:1) into the Harbor, in a very short time, eliminates any concern about IDOD effects in the receiving water.

## RESULTS AND DISCUSSION

The results for the February 2007 bioassay tests are included in Attachment II. The 96-hour $LC_{50}$ for the effluent tested was 44.1% percent effluent. The no observable effects concentration (NOEC) for the 96-hour bioassay was 25% effluent, and the least observable effects concentration (LOEC) was 50% percent. Results on a daily basis are summarized in Table 2.

**Table 2**
**StarKist Samoa and COS Samoa Packing**
**Combined Effluent Bioassay Results**
**February 2007 Sampling**

| Exposure Time | Parameter | | |
|---|---|---|---|
| | $LC_{50}$ | NOEC | LOEC |
| 24 hours | 64.9% | 50% | 75% |
| 48 hours | 57.2% | 25% | 50% |
| 72 hours | 49.9% | 25% | 50% |
| 96 hours | 44.1% | 25% | 50% |

### Comparison to Previous Tests

Table 3 summarizes the results of the effluent bioassay tests for the samples collected in the February 2007 sampling compared to the previous bioassay tests. Figure 1 summarizes the $LC_{50}$ for the mysid and penaeid tests done since February 1993. Figure 2 presents the range of $LC_{50}$ results for mysids tests conducted since 1994. The $LC_{50}$, NOEC, and LOEC are within the range obtained from previous tests where *Americamysis bahia* (*Mysidopsis bahia*) was used in place of *Penaeus vannami*.

### Conclusions

The bioassay tests for the Joint Cannery Outfall effluent for February 2007 indicate that effluent toxicity levels are not of concern. The time scale of the mixing of the effluent with the receiving water is on the order of seconds to achieve dilutions that will eliminate possible toxic effects as reflected by the bioassay results. The discharge is located in about 180 feet of water and the effluent toxicity tests indicate that the discharge is diluted to non-toxic levels immediately after discharge and well within the initial dilution plume.

For the February 2007 test the $LC_{50}$ of 44.1 percent corresponds to 2.27 acute toxicity units ($TU_a$). A dilution of less than 8:1 will reduce the toxicity to less than 0.3 $TU_a$, which is considered the acceptable level for the protection of aquatic life. The JCO achieves an initial dilution, under critical conditions of greater than 300:1. Therefore, at the edge of the zone of initial dilution (ZID) the acute toxicity is less than 0.01 $TU_a$ for the $LC_{50}$ documented in the February test.

## Table 3
## StarKist Samoa and COS Samoa Packing
## Combined Effluent Bioassay Results

| Date | *Species* | Parameters | | |
|---|---|---|---|---|
| | | $LC_{50}$ | NOEC | LOEC |
| 2/93 | *Penaeus vannami* | 4.8%[1] | 3.1% | 6.25% |
| 10/93 | *Penaeus vannami* | 15.67% | 3.1% | 6.25% |
| 2/94 | *Penaeus vannami* | 15.76% | <1.6% | 1.6% |
| 10/94 | *Mysidopsis bahia*[2] | 31.2% | 25% | 50% |
| 3/95 | *Penaeus vannami* | 14.8% | 6.25% | 12.5% |
| 3/95 | *Mysidopsis bahia*[3] | 10.8% | 6.25% | 12.5% |
| 2/96 | *Penaeus vannami* | >50% | >50% | >50% |
| 2/96 | *Mysidopsis bahia*[3] | 28.36% | 12.5% | 25% |
| 3/96 | *Penaeus vannami* | 44.4% | 25% | 50% |
| 11/96 | *Penaeus vannami* | 7.11% | 3.1% | 6.25% |
| 03/97 | *Penaeus vannami* | 39.36% | 12.5% | 25% |
| 09/97 | *Penaeus vannami*[4] | 12.3% | 6.25% | 12.5% |
| 06/98 | *Mysidopsis bahia*[2] | 17.2% | 6.25% | 12.5% |
| 11/98 | *Mysidopsis bahia*[2] | 15% | 6.25% | 12.5% |
| 02/00 | *Mysidopsis bahia*[2] | 20% | 6.25% | 12.5% |
| 08/00 | *Mysidopsis bahia*[2] | 17.1% | 3.1% | 6.25% |
| 03/01 | *Americamysis bahia*[2,5] | 13.8% | 12.5% | 25% |
| 10/01 | *Americamysis bahia*[2,6] | 37.5% | 25% | 50% |
| 3/02 | *Americamysis bahia*[2,6] | 16.1% | 12.5% | 25% |
| 8/02 | *Americamysis bahia*[2,6] | 10.23% | 6.25% | 12.5% |
| 03/03 | *Americamysis bahia*[2,6] | 28.4% | 25% | 50% |
| 08/03 | *Americamysis bahia*[2,6] | 43.2% | 25% | 50% |
| 02/04 | *Americamysis bahia*[2,6] | >50% | 50% | >50% |
| 09/04 | *Americamysis bahia*[2,6] | >50% | 50% | >50% |
| 03/05 | *Americamysis bahia*[2,6] | 48.5% | 25% | 50% |
| 08/05 | *Americamysis bahia*[2,6] | >50% | 50% | >50% |
| 03/06 | *Americamysis bahia*[2,6] | 35.6%[7] | 25% | 50% |
| 05/06 | *Americamysis bahia*[2,6] | 32.7%[7] | 12.5% | 25% |
| 11/06 | *Americamysis bahia*[2,6] | 43.1% | 25% | 50% |
| **02/07** | ***Americamysis bahia***[2,6] | **44.1%** | **25%** | **50%** |

[1]The February 1993 samples were not aerated until after the first day of the test. For subsequent tests the samples were aerated for the entire duration of the tests.
[2]*Mysidopsis bahia* used as substitutes because *Penaeus vannami* not available: as directed and approved by USEPA.
[3]*Mysidopsis bahia* used in addition to *Penaeus vannami* as described in text of technical memorandums reporting test results. Only one species is required by the permit conditions.
[4]Stage 1 (3 mm) *Penaeus vannami* were used for testing because older Stage 7 and 8 (8-10 mm) *Penaeus vannami* were not available.
[5]*Mysidopsis bahia* renamed *Americamysis bahia*. Results indicate increased toxicity because of low DO in renewal concentrations as renewal water was not aerated prior to use
[6] *Mysidopsis bahia* renamed *Americamysis bahia*
[7] Results for this test depressed because aeration was not provided (see text).

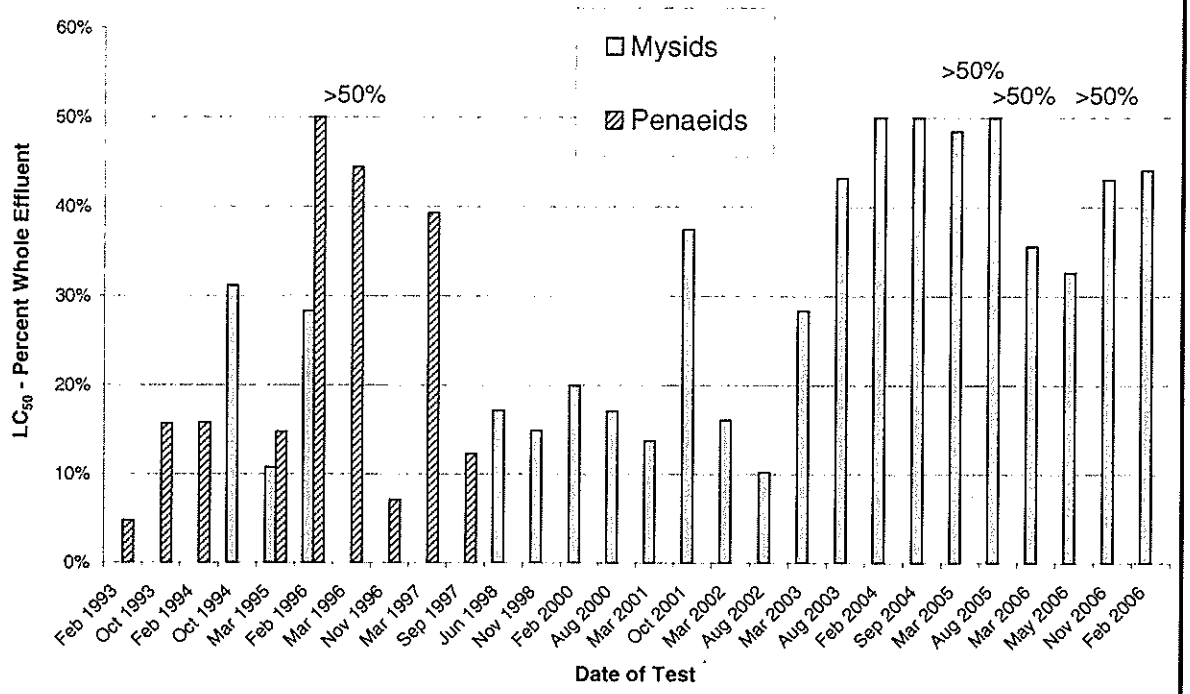


**Figure 1. $LC_{50}$ for Mysid and Penaeid Tests**

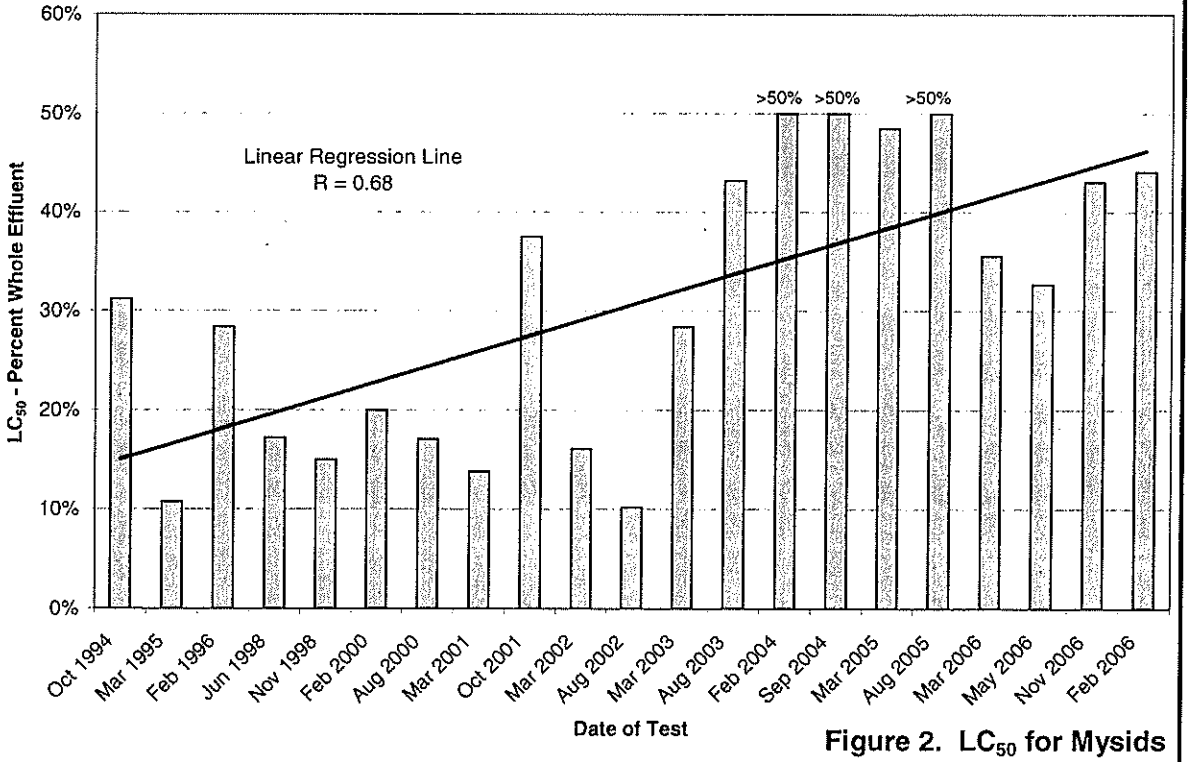


**Figure 2. $LC_{50}$ for Mysids**

# ATTACHMENT I

## Chain-of-Custody

# CHAIN OF CUSTODY

| | | | | | | |
|---|---|---|---|---|---|---|
| PROJECT: | Joint Cannery Outfall Effluent Monitoring- JCO0701.NT | | | | | |
| FROM: | Karen Glatzel, gdc | | | | | |
| | P.O. Box 1238, Trinidad, CA, 95570-1238 | | | | 707-677-0123 | gdcocn@eathlink.net |
| TO: | Brian Buzby, EnviroSystems, Inc. | | | | | |
| | One Lafayette Road, Hampton, New Hampshire 03842 | | | | 603-926-3345 | |

| SAMPLE I.D. | DATE | TIME | MATRIX | NUMBER OF CONTAINERS | ANALYSIS REQUESTED<br>96-hour acute mysid bioassay test | COMMENTS |
|---|---|---|---|---|---|---|
| JCO-07NT | 2/28/2007 | | Water | 1 | X | |
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|---|---|---|---|
| SAMPLED BY: K. Glatzel | DATE/TIME: | 2/28/2007 | SPECIAL INSTRUCTIONS/REMARKS: |
| SHIPPED VIA: DHL | DATE/TIME: | 3/1/2007 | Note - sample may be approximately 40% seawater |
| RELINQUISHED BY: S.Costa | DATE/TIME: | 3/1/2007 | **Dilutions and aeration protocols as in previous test** |
| RECEIVED BY: | DATE/TIME: | | |
| RELINQUISHED BY: | DATE/TIME: | | |
| RECEIVED BY: | DATE/TIME: | | |

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Company Name: STARKIST SAMOA

Address: P.O. Box 368

PAGO PAGO, AMERICAN SAMOA

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|---|---|
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Contact Person: BRIAN BUZBY

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|---|---|---|---|---|---|
| 1 | 6 lbs | ____@ | | x | x |
| | | ____@ | | x | x |
| | | ____@ | | x | x |

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FOR LABORATORY ANALYSIS

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# ATTACHMENT II

EnviroSystems, Inc. Laboratory Report
for February 2007 Sampling

# TOXICOLOGICAL EVALUATION OF A TREATED EFFLUENT: BIOMONITORING SUPPORT FOR A NPDES PERMIT March 2007

**American Samoa Joint Cannery Outfall**

Prepared For

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March 2007
Reference Number CH2M-Samoa15566-07-03

## STUDY NUMBER 15566

## EXECUTIVE SUMMARY

The following summarizes the results of acute exposure bioassays performed from March 6-10, 2007 in support of the NPDES biomonitoring requirements of the American Samoa Joint Cannery Outfall. The 96 hour acute definitive assay was conducted using the marine species, *Americamysis bahia.*

The salinity adjusted effluent sample collected from the American Samoa Joint Cannery Outfall exhibited significant signs of acute toxicity to the mysid shrimp, *Americamysis bahia,* during the 96 hour exposure period.

**Acute Toxicity Evaluation**

| Species | Exposure | LC-50 | NOEC | LOEC |
|---|---|---|---|---|
| *Americamysis bahia* | 24-Hours | 64.9% | 50% | 75% |
| | 48-Hours | 57.2% | 25% | 50% |
| | 72-Hours | 49.9% | 25% | 50% |
| | 96-Hours | 44.1% | 25% | 50% |

# TOXICOLOGICAL EVALUATION
# OF A TREATED EFFLUENT:
# BIOMONITORING SUPPORT FOR A NPDES PERMIT
# March 2007

## American Samoa Joint Cannery Outfall

## 1.0 INTRODUCTION

This report presents the results of an acute toxicity test conducted on an effluent sample collected from the American Samoa Joint Cannery Outfall. Testing was based on programs and protocols developed by the US EPA (2002) and involved conducting 96 hour acute static renewal toxicity tests with the marine species, *Americamysis bahia*. Testing was performed at EnviroSystems, Incorporated (ESI), Hampton, New Hampshire in accordance with the provisions of the NELAC Standards (2000).

Acute toxicity tests involve preparing a series of concentrations by diluting effluent with control water. Groups of test organisms are exposed to each effluent concentration and a control for a specified period. In acute tests, mortality data for each concentration are used to calculate (by regression) the median lethal concentration, or LC-50, defined as the effluent concentration which kills half of the test organisms. Samples with high LC-50 values are less likely to cause significant environmental impact. The acute no observed effect concentration (NOEC) and lowest observed effect concentration (LOEC) document the highest and lowest effluent concentrations that have no impact and a significant impact on the test species, respectively.

## 2.0 MATERIALS AND METHODS

### 2.1 General Methods

Toxicological and analytical protocols used in this program follow procedures primarily designed by the EPA to provide standard approaches for the evaluation of toxicological effects of discharges on aquatic organisms, and for the analysis of water samples. See Section 4.0 for a list of references.

### 2.2 Test Species

Every attempt was made to acquire the species, *Penaeus vannami,* as this is the preferred organism under the Cannery's permit. ESI was unable to obtain reasonably priced *P. vannami*. Due to the exorbitant expense, the decision was made to use an alternate species, *Americamysis bahia*.

*A. bahia*, ≤5 days old, were obtained from Aquatic Research Organisms, Hampton, New Hampshire. Test organisms were transferred to test chambers by large bore pipet, minimizing the amount of water added to test solutions.

2.3 Effluent and Dilution Water

The effluent sample used in the assay was identified as "JC0-07NT". Sample collection information is provided in Table 1. Upon receipt, the sample was stored at 4°C. All sample material used in the assay was warmed to 20±1°C prior to preparing test solutions. Total residual chlorine (TRC) was measured using amperometric titration (MDL 0.05 mg/L). As the effluent sample contained <0.05 mg/L, TRC dechlorination with sodium thiosulfate was not required (EPA 2002). Aliquots of the undiluted effluent sample were collected for ammonia analysis when the sample arrived and again prior to renewal. Upon arrival, the effluent sample had a salinity of 9.0‰. Salinity of the effluent was increased to 25‰ by the addition of artificial sea salts. Test concentrations for the assays were 100%, 75%, 50%, 25%, 12.5%, and 6.25% effluent with a laboratory water diluent control.

The dilution water used in this assay was collected from the sea water system at ESI. The water is pumped in daily from the Hampton Estuary on the flood tide, filtered through a high volume sand filter, and stored in 3000 gallon polyethylene tanks. The water is classified as Class SA-1 by the State of New Hampshire, and has been used to culture test organisms for over 20 years. Sea water used in the assay had a salinity of 25±2‰ and a TRC of <0.05 mg/L.

2.4 Acute Toxicity Tests

The 96 hour acute static renewal toxicity test was conducted at 20±2°C with a photoperiod of 16:8 hours light:dark. Test chambers for the acute assays were 250 mL glass beakers containing 200 mL test solution in each of 5 replicates, with 10 organisms/replicate. Survival, dissolved oxygen, pH, salinity and temperature were measured daily in all replicates. Test solutions were renewed after 48 hours using effluent from the start sample. Mysid shrimp were fed daily with <24 hour old brine shrimp.

2.5 Data Analysis

Survival data were analyzed at 24 hour intervals to assess toxicity using CETIS, Comprehensive Environmental Toxicity Testing System, software. The program computes acute exposure endpoints based on EPA decision tree guidelines specified in individual test methods. For acute exposure endpoints statistical significance was accepted at $\propto$ <0.05.

2.6 Quality Control

As part of the laboratory quality control program, standard reference toxicant assays are conducted on a regular basis for each test species. These results provide relative health and response data while allowing for comparison with historic data sets. See Table 2 for details.

## 3.0 RESULTS

Results of the acute exposure bioassay conducted using the mysid shrimp, *A. bahia*, are summarized in Tables 3A and 3B. Effluent and dilution water characteristics are presented in Table 4. Table 5 provides a summary of historic data associated with the discharge. Support data are included in Appendix A.

Minimum test acceptability criteria require ≥90% survival in the control concentration. As the laboratory water diluent control met or exceeded this protocol specification, results associated with the assay indicate healthy test organisms were used and that the dilution water had no adverse impact on the outcome of the assay. These data are considered as valid for evaluating impacts associated with the effluent sample.

Table 3 provides a summary of the acute exposure data and results.

## 4.0 LITERATURE CITED


APHA. 1998. *Standard Methods for the Examination of Water and Wastewater*, 20th Edition. Washington D.C.

*National Environmental Laboratory Accreditation Conference*: Quality Systems. Chapter 5. June 2000.

Stephan, C. 1982. Documentation for Computing LC-50 Values with a Mini Computer. Unpublished.

US EPA. 2002. *Attachment G: NPDES Whole Effluent Toxicity Testing, Monitoring and Reporting Tips and Common Pitfalls.* Dated December 2002. US EPA Region I Offices, Boston, Massachusetts.

U.S. EPA. 2002. *Methods for Measuring the Acute Toxicity of Effluents to Freshwater and Marine Organisms.* Fifth Edition. EPA-821-R-02-012.

**TABLE 1. Summary of Sample Collection Information. American Samoa Joint Cannery Outfall Effluent Evaluation. March 2007.**

| Sample Description | Type | Collection Date | Collection Time | Receipt Date | Receipt Time | Arrival Temp °C |
|---|---|---|---|---|---|---|
| EFFLUENT | Comp | 02/28/07 | No data | 03/06/07 | 0930 | 12* |

* Upon receipt, the temperature was outside of the range of 4±2°C recommended by the protocol.

**TABLE 2. Summary of Reference Toxicant Data. American Samoa Joint Cannery Outfall Effluent Evaluation. March 2007.**

| Date | Endpoint | | Value | Historic Mean/ Central Tendency | Acceptable Range | Reference Toxicant |
|---|---|---|---|---|---|---|
| *A. bahia* | | | | | | |
| 02/20/07 | Survival | LC-50 | 21.9 | 20.8 | 15.4 - 26.2 | SDS (mg/L) |

Means and Acceptable Ranges based on the most recent 20 reference toxicant assays

**TABLE 3A. Summary of Acute Evaluation Results.**
**American Samoa Joint Cannery Outfall Effluent Evaluation.**
**March 2007.**

| Concentration % Effluent | Exposure | Replicates A | B | C | D | E | Mean | Standard Deviation | Coefficient of Variation |
|---|---|---|---|---|---|---|---|---|---|
| Lab Control | Start | 10 | 10 | 10 | 10 | 10 | 100% | 0.000 | 0.00% |
| | 24-Hours | 9 | 10 | 10 | 9 | 10 | 96% | 0.490 | 5.10% |
| | 48-Hours | 9 | 10 | 10 | 9 | 10 | 96% | 0.490 | 5.10% |
| | 72 Hours | 9 | 10 | 10 | 9 | 10 | 96% | 0.490 | 5.10% |
| | 96-Hours | 9 | 10 | 10 | 9 | 10 | 96% | 0.490 | 5.10% |
| 6.25% | 24-Hours | 9 | 10 | 9 | 10 | 10 | 96% | 0.490 | 5.10% |
| | 48-Hours | 9 | 10 | 9 | 10 | 9 | 94% | 0.490 | 5.21% |
| | 72 Hours | 9 | 9 | 8 | 9 | 8 | 86% | 0.490 | 5.70% |
| | 96-Hours | 9 | 9 | 8 | 9 | 8 | 86% | 0.490 | 5.70% |
| 12.5% | 24-Hours | 10 | 10 | 9 | 10 | 10 | 98% | 0.400 | 4.08% |
| | 48-Hours | 10 | 10 | 7 | 10 | 9 | 92% | 1.166 | 12.68% |
| | 72 Hours | 10 | 10 | 7 | 6 | 6 | 78% | 1.833 | 23.50% |
| | 96-Hours | 10 | 10 | 7 | 6 | 6 | 78% | 1.833 | 23.50% |
| 25% | 24-Hours | 10 | 10 | 10 | 10 | 9 | 98% | 0.400 | 4.08% |
| | 48-Hours | 10 | 10 | 10 | 10 | 9 | 98% | 0.400 | 4.08% |
| | 72 Hours | 9 | 8 | 9 | 9 | 9 | 88% | 0.400 | 4.55% |
| | 96-Hours | 9 | 8 | 5 | 9 | 9 | 80% | 1.549 | 19.36% |
| 50% | 24-Hours | 7 | 10 | 7 | 10 | 8 | 84% | 1.356 | 16.15% |
| | 48-Hours | 7 | 6 | 6 | 10 | 6 | 70% | 1.549 | 22.13% |
| | 72 Hours | 5 | 4 | 1 | 6 | 5 | 42% | 1.720 | 40.96% |
| | 96-Hours | 2 | 4 | 1 | 6 | 5 | 36% | 1.855 | 51.52% |
| 75% | 24-Hours | 0 | 2 | 5 | 4 | 0 | 22% | 2.040 | 92.71% |
| | 48-Hours | 0 | 2 | 2 | 1 | 0 | 10% | 0.894 | 89.44% |
| | 72 Hours | 0 | 2 | 2 | 0 | 0 | 8% | 0.980 | 122.47% |
| | 96-Hours | 0 | 0 | 0 | 0 | 0 | 0% | 0.000 | ?? |
| 100% | 24-Hours | 1 | 2 | 0 | 1 | 0 | 8% | 0.748 | 93.54% |
| | 48-Hours | 0 | 0 | 0 | 0 | 0 | 0% | 0.000 | ?? |
| | 72 Hours | 0 | 0 | 0 | 0 | 0 | 0% | 0.000 | ?? |
| | 96-Hours | 0 | 0 | 0 | 0 | 0 | 0% | 0.000 | ?? |

**TABLE 3B. Summary of Acute Evaluation Results. American Samoa Joint Cannery Outfall Effluent Evaluation. March 2007.**

SUMMARY OF ENDPOINTS

| Exposure Period | LC-50 (95% Limits) | METHOD | NOEC | LOEC |
|---|---|---|---|---|
| 24 Hours | 64.9% (59.8-69.6) | Dunnett's Multiple Comparison | 50% | 75% |
| 48 Hours | 57.2% (53.1-61.1) | Dunnett's Multiple Comparison | 25% | 50% |
| 72 Hours | 49.9% (42.7-54.8) | Dunnett's Multiple Comparison | 25% | 50% |
| 96 Hours | 44.1% (26.6-58.9) | Dunnett's Multiple Comparison | 25% | 50% |

**TABLE 4. Summary of Effluent and Diluent Characteristics. American Samoa Joint Cannery Outfall Effluent Evaluation. March 2007.**

| PARAMETER | UNITS | 100% EFFLUENT | 50% EFFLUENT | DILUENT |
|---|---|---|---|---|
| Salinity - As Received | ‰ | 9.0 | - | 25 |
| Salinity - After Salinity Adjustment | ‰ | 25 | 25 | - |
| pH - As Received | SU | 6.97 | - | 7.97 |
| pH - After Salinity Adjustment | SU | 7.44 | 7.62 | - |
| TRC - As Received | mg/L | <0.05 | - | <0.05 |
| Dissolved Oxygen - As Received | mg/L | 0.7 | - | 7.4 |
| Dissolved Oxygen - After Aeration | mg/L | 5.0 | 5.6 | - |
| Ammonia - As Received | mg/L as N | 28 | - | <0.1 |
| Unionized Ammonia - As Received | mg/L as N | 0.103 | - | 0.004 |
| Ammonia - Salinity Adjusted | mg/L as N | - | 15 | - |
| Unionized Ammonia - Salinity Adjusted | mg/L as N | - | 0.243 | - |
| Ammonia - at 48 Hours | mg/L as N | 2.4 | 1.9 | <0.1 |
| Unionized Ammonia - at 48 Hours | mg/L as N | 0.165 | 0.084 | 0.002 |

**TABLE 5. Summary of StarKist Samoa and COS Samoa Packing Combined Effluent Bioassay Results. American Samoa Joint Cannery Outfall Effluent Evaluation. March 2007.**

| Date | Species | 96-Hour Endpoints | | |
|---|---|---|---|---|
| | | LC-50 | NOEC | LOEC |
| 02/93[1] | *Penaeus vannami* | 4.8% | 3.1% | 6.25% |
| 10/93[1] | *Penaeus vannami* | 15.67% | 3.1% | 6.25% |
| 02/94[1] | *Penaeus vannami* | 15.76% | <1.6% | 1.6% |
| 10/94[1] | *Americamysis bahia* | 31.2% | 25.0% | 50.0% |
| 03/95[1] | *Penaeus vannami* | 14.8% | 6.25% | 12.5% |
| 03/95[1] | *Americamysis bahia* | 10.8% | 6.25% | 12.5% |
| 02/96[1] | *Penaeus vannami* | >50.0% | >50.0% | >50.0% |
| 03/96[1] | *Penaeus vannami* | 44.4% | 25.0% | 50.0% |
| 11/96[1] | *Penaeus vannami* | 7.11% | 3.1% | 6.25% |
| 03/97[1] | *Penaeus vannami* | 39.36% | 12.5% | 25.0% |
| 09/97[1] | *Penaeus vannami* | 12.3% | 6.25% | 12.5% |
| 06/98[1] | *Americamysis bahia* | 17.2% | 6.25% | 12.5% |
| 11/98[1] | *Americamysis bahia* | 15.0% | 6.25% | 12.5% |
| 02/00[1] | *Americamysis bahia* | 20.0% | 6.25% | 12.5% |
| 08/00[1] | *Americamysis bahia* | 17.1% | 3.1% | 6.25% |
| 03/01[2] | *Americamysis bahia* | 13.81% | 12.5% | 25.0% |
| 03/02[2] | *Americamysis bahia* | 16.13% | 12.5% | 25.0% |
| 08/02[2] | *Americamysis bahia* | 10.23% | 6.25% | 12.5% |
| 03/03[2] | *Americamysis bahia* | 28.4% | 25.0% | 50.0% |
| 08/03[2] | *Americamysis bahia* | 43.2% | 25.0% | 50.0% |
| 03/04[2] | *Americamysis bahia* | >50.0% | 50.0% | >50.0% |
| 10/04[2] | *Americamysis bahia* | >50.0% | 50.0% | >50.0% |
| 03/05[2] | *Americamysis bahia* | 48.5% | 25.0% | 50.0% |
| 10/05[2] | *Americamysis bahia* | >50.0% | 50.0% | >50.0 |
| 03/06[2] | *Americamysis bahia* | 36.6% | 25% | 50% |
| 11/06[2] | *Americamysis bahia* | 43.1% | 25% | 50% |
| 03/07[2] | *Americamysis bahia* | 44.1% | 25% | 50% |

Notes:

1. Assays conducted by Advanced Biological Testing, Inc., Rohnert Park, California
2. Assays conducted by EnviroSystems, Inc., Hampton, New Hampshire

## APPENDIX A

## DATA SHEETS

## STATISTICAL SUPPORT

| Contents | Number of Pages |
|---|---|
| Methods Used in NPDES Permit Biomonitoring Testing | 1 |
| *A. bahia* Acute Bioassay Data Summary | 3 |
| *A. bahia* Survival Statistics: LC-50, NOEC | 19 |
| *A. bahia* Organism Culture Sheet | 1 |
| Preparation of Dilutions | 1 |
| Record of Meters Used for Water Quality Measurements | 1 |
| Unionized Ammonia Calculation | 3 |
| Sample Receipt Record | 1 |
| Chain of Custody | 1 |
| Total Appendix Pages | 31 |

## METHODS USED IN NPDES PERMIT BIOMONITORING TESTING

| Parameter | Method |
|---|---|
| **Acute Exposure Bioassays** | |
| *Ceriodaphnia dubia, Daphnia pulex* | EPA-821-R-02-012 |
| *Pimephales promelas* | EPA-821-R-02-012 |
| *Americamysis bahia* | EPA-821-R-02-012 |
| *Menidia beryllina, Cyprinodon variegatus* | EPA-821-R-02-012 |
| **Chronic Exposure Bioassays** | |
| *Ceriodaphnia dubia* | EPA-821-R-02-013, 1002.0 |
| *Pimephales promelas* | EPA-821-R-02-013, 1000.0 |
| *Cyprinodon variegatus* | EPA-821-R-02-014, 1004.0 |
| *Menidia beryllina* | EPA-821-R-02-014, 1006.0 |
| *Arbacia punctulata* | EPA-821-R-02-014, 1008.0 |
| *Champia parvula* | EPA-821-R-02-014, 1009.0 |
| **Trace Metals:** | |
| ICP Metals | EPA 200.7/SW 6010 |
| Hardness | Standard Methods 20th Edition - Method 2340 B |
| **Wet Chemistries:** | |
| Alkalinity | EPA 310.2 |
| Chlorine, Residual | Standard Methods 20th Edition - Method 4500CLD |
| Total Organic Carbon | Standard Methods 20th Edition - Method 5310C |
| Specific Conductance | Standard Methods 20th Edition - Method 2510B |
| Nitrogen - Ammonia | Standard Methods 20th Edition - Method 4500NH3G |
| pH | Standard Methods 20th Edition - Method 4500H+B |
| Solids, Total (TS) | Standard Methods 20th Edition - Method 2540.B |
| Solids, Total Suspended (TSS) | Standard Methods 20th Edition - Method 2540D |
| Dissolved Oxygen | Standard Methods 20th Edition - Method 4500-O G |

# ACUTE BIOASSAY DATA SUMMARY

| STUDY: 15566 | | "AS RECEIVED" EFFLUENT AND DILUENT CHEMISTRIES | | | | | |
|---|---|---|---|---|---|---|---|
| CLIENT: CH2M Hill | TEST ORGANISM: *A. bahia* | | TRC | AMM 0 HR✖ | AMM 48 HR✖ | pH | DO | Salinity |
| SAMPLE: American Samoa | ORGANISM SUPPLIER/BATCH/AGE: *See Organism Culture Sheet* | EFFLUENT | See "EFFLUENT & DILUENT CHEMISTRY and WATER QUALITY DATA" sheet | | | | | |
| DILUENT: LAB SALT | | DILUENT | | | | | | |

SALINITY ADJUSTMENT RECORD (IF APPLICABLE): 8000 ML EFFLUENT + 147 G SEA SALTS = 100% ACTUAL PERCENTAGE

| CONC | REP | SURVIVAL | | | | | ✦DISSOLVED OXYGEN (MG/L)✦ | | | | | | PH (SU) | | | | | | TEMPERATURE (°C) | | | | | | SALINITY (ppt) | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | 0 | 24 | 48 | 72 | 96 | 0 | 24 | 48◊ | 48☆ | 72 | 96 | 0 | 24 | 48◊ | 48☆ | 72 | 96 | 0 | 24 | 48◊ | 48 | 72 | 96 | 0 | 24 | 48◊ | 48 | 72 | 96 |
| LAB | A | 10 | 9 | 9 | 9 | 9 | 7.4 | 7.1 | 7.4 | 7.3 | 6.0 | 6.4 | 7.97 | 7.76 | 7.73 | 7.59 | 7.67 | 7.69 | 20 | 19 | 19 | 20 | 19 | 20 | 25 | 26 | 27 | 26 | 26 | 26 |
| | B | 10 | 10 | 10 | 10 | 10 | 7.4 | 7.1 | 7.3 | 7.3 | 6.0 | 6.4 | 7.97 | 7.79 | 7.84 | 7.70 | 7.68 | 7.70 | 20 | 19 | 19 | 20 | 19 | 20 | 25 | 26 | 27 | 26 | 27 | 27 |
| | C | 10 | 10 | 10 | 10 | 10 | 7.4 | 7.1 | 7.8 | 7.3 | 6.0 | 6.4 | 7.98 | 7.79 | 7.86 | 7.73 | 7.70 | 7.70 | 20 | 19 | 19 | 20 | 19 | 20 | 25 | 26 | 28 | 26 | 26 | 27 |
| | D | 10 | 9 | 9 | 9 | 9 | 7.4 | 7.1 | 7.6 | 7.3 | 6.0 | 6.4 | 7.98 | 7.79 | 7.76 | 7.68 | 7.68 | 7.68 | 20 | 19 | 19 | 20 | 19 | 20 | 25 | 26 | 27 | 26 | 26 | 27 |
| | E | 10 | 10 | 10 | 10 | 10 | 7.4 | 7.1 | 7.9 | 7.3 | 6.5 | 6.4 | 7.98 | 7.79 | 7.87 | 7.70 | 7.71 | 7.69 | 20 | 19 | 19 | 20 | 19 | 20 | 25 | 26 | 29 | 26 | 27 | 27 |
| 6.25% | A | 10 | 9 | 9 | 9 | 9 | 7.0 | 7.2 | 7.3 | 7.3 | 6.3 | 6.3 | 7.95 | 7.74 | 7.81 | 7.76 | 7.80 | 7.71 | 20 | 19 | 19 | 20 | 19 | 20 | 25 | 26 | 27 | 26 | 26 | 27 |
| | B | 10 | 10 | 10 | 9 | 9 | 7.0 | 7.1 | 7.2 | 7.3 | 6.3 | 6.2 | 7.95 | 7.74 | 7.60 | 7.74 | 7.74 | 7.72 | 20 | 19 | 19 | 20 | 19 | 20 | 25 | 26 | 27 | 25 | 27 | 27 |
| | C | 10 | 9 | 9 | 8 | 8 | 7.0 | 7.1 | 7.0 | 7.3 | 6.0 | 6.2 | 7.94 | 7.74 | 7.81 | 7.76 | 7.74 | 7.71 | 20 | 19 | 19 | 20 | 19 | 20 | 25 | 26 | 27 | 26 | 27 | 27 |
| | D | 10 | 10 | 10 | 9 | 9 | 7.0 | 7.1 | 7.3 | 7.3 | 6.0 | 6.2 | 7.95 | 7.83 | 7.83 | 7.78 | 7.74 | 7.69 | 20 | 19 | 19 | 20 | 19 | 20 | 25 | 26 | 28 | 26 | 27 | 27 |
| | E | 10 | 10 | 9 | 8 | 8 | 7.0 | 7.1 | 7.4 | 7.3 | 6.0 | 6.2 | 7.96 | 7.86 | 7.88 | 7.79 | 7.74 | 7.70 | 20 | 19 | 19 | 20 | 19 | 20 | 25 | 26 | 29 | 26 | 27 | 27 |
| DATE | | 3/6 | 3/7 | 3/8 | 3/9 | 3/10 | 3/6/07 | 3/7/07 | 3/8 | 3/8 | 3/9 | 3/10 | | | | | | | | | | | | | | | | | | |
| TIME | | 1600 | 1500 | 1530 | 1500 | 1400 | 1545 | 1515 | 1455 | 1600 | 1600 | 1500 | | | | | | | | | | | | | | | | | | |
| INITIALS | | SJ | MPB | CS | CS | CS | SJ | CS | NT | CS | CS | CS | | | | | | | | | | | | | | | | | | |
| FED? | | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | | | | | | | | | | | | | | | | | | |

✖ - See:" EFFLUENT & DILUENT CHEMISTRY and WATER QUALITY DATA" sheet.
✦ -AERATE PRIOR TO MIXING DILUTIONS - AERATE TEST CHAMBERS FROM START!
◊ - "Old" water qualities (prior to renewal) ☆ - "New"water qualities (post renewal)

# ACUTE BIOASSAY DATA SUMMARY

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| STUDY: 15566 | SAMPLE RECEIVED: | "AS RECEIVED" EFFLUENT AND DILUENT CHEMISTRIES | | | | | |
| CLIENT: CH2M Hill | TEST ORGANISM: *A. bahia* | | TRC | AMM 0 HR✖ | AMM 48 HR✖ | pH | DO | Salinity |
| SAMPLE: American Samoa | ORGANISM SUPPLIER: | EFFLUENT | See "EFFLUENT & DILUENT CHEMISTRY and WATER QUALITY DATA" sheet | | | | | |
| DILUENT: LAB SALT | ORGANISM BATCH/AGE: | DILUENT | | | | | | |

| CONC | REP | SURVIVAL | | | | | ✦DISSOLVED OXYGEN (MG/L)✦ | | | | | | PH (SU) | | | | | | TEMPERATURE (°C) | | | | | | SALINITY (ppt) | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | 0 | 24 | 48 | 72 | 96 | 0 | 24 | 48◊ | 48☆ | 72 | 96 | 0 | 24 | 48◊ | 48☆ | 72 | 96 | 0 | 24 | 48◊ | 48 | 72 | 96 | 0 | 24 | 48◊ | 48 | 72 | 96 |
| 12.5% | A | 10 | 10 | 10 | 9 | 10 | 6.8 | 7.2 | 7.7 | 7.3 | 7.3 | 6.2 | 7.93 | 7.86 | 7.97 | 7.80 | 7.67 | 7.60 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 27 | 26 | 26 | 27 |
| | B | 10 | 10 | 10 | 10 | 10 | 6.8 | 7.2 | 7.1 | 7.3 | 7.2 | 6.0 | 7.92 | 7.86 | 7.81 | 7.76 | 7.68 | 7.70 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 28 | 26 | 26 | 27 |
| | C | 10 | 9 | 7 | 7 | 7 | 6.8 | 7.1 | 7.2 | 7.3 | 7.2 | 6.0 | 7.92 | 7.79 | 7.90 | 7.76 | 7.70 | 7.71 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 27 | 26 | 26 | 27 |
| | D | 10 | 10 | 10 | 6 | 6 | 6.8 | 7.1 | 7.3 | 7.3 | 7.2 | 6.2 | 7.92 | 7.84 | 7.88 | 7.78 | 7.70 | 7.69 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 29 | 26 | 26 | 27 |
| | E | 10 | 10 | 9 | 6 | 6 | 6.8 | 7.1 | 7.3 | 7.3 | 7.2 | 6.2 | 7.92 | 7.87 | 7.86 | 7.78 | 7.71 | 7.68 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 29 | 26 | 26 | 27 |
| 25% | A | 10 | 10 | 10 | 9 | 9 | 6.2 | 6.9 | 7.4 | 7.3 | 7.2 | 6.2 | 7.80 | 7.83 | 8.01 | 7.80 | 7.80 | 7.74 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 27 | 26 | 26 | 27 |
| | B | 10 | 10 | 10 | 8 | 8 | 6.2 | 7.0 | 7.0 | 7.3 | 7.2 | 6.0 | 7.80 | 7.89 | 7.94 | 7.79 | 7.74 | 7.74 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 27 | 26 | 26 | 27 |
| | C | 10 | 10 | 10 | 9 | 5 | 6.2 | 6.9 | 7.2 | 7.3 | 7.3 | 6.0 | 7.81 | 7.90 | 7.89 | 7.79 | 7.75 | 7.70 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 27 | 26 | 27 | 27 |
| | D | 10 | 10 | 10 | 9 | 9 | 6.2 | 6.9 | 6.5 | 6.5 | 6.2 | 6.0 | 7.81 | 7.90 | 7.91 | 7.80 | 7.81 | 7.81 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 28 | 26 | 27 | 27 |
| | E | 10 | 8 | 8 | 9 | 9 | 6.2 | 7.0 | 7.1 | 6.5 | 6.3 | 6.0 | 7.81 | 7.90 | 7.92 | 7.81 | 7.70 | 7.81 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 30 | 26 | 27 | 27 |
| 50% | A | 10 | 7 | 7 | 5 | 2 | 5.6 | 6.5 | 6.6 | 6.5 | 6.3 | 6.0 | 7.62 | 7.84 | 8.07 | 7.78 | 7.87 | 7.80 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 27 | 26 | 27 | 27 |
| | B | 10 | 10 | 6 | 4 | 4 | 5.6 | 6.3 | 6.2 | 6.4 | 6.2 | 5.9 | 7.62 | 7.84 | 8.02 | 7.79 | 8.01 | 8.02 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 28 | 26 | 27 | 27 |
| | C | 10 | 7 | 6 | 1 | 1 | 5.6 | 6.3 | 6.2 | 6.4 | 6.4 | 6.4 | 7.63 | 7.89 | 8.07 | 7.82 | 7.87 | 7.90 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 28 | 26 | 28 | 28 |
| | D | 10 | 10 | 10 | 6 | 6 | 5.6 | 6.3 | 6.1 | 6.2 | 6.2 | 6.2 | 7.63 | 7.95 | 8.07 | 7.92 | 7.88 | 7.90 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 28 | 24 | 27 | 28 |
| | E | 10 | 8 | 6 | 5 | 5 | 5.6 | 6.3 | 6.3 | 6.2 | 6.0 | 6.2 | 7.63 | 7.97 | 8.04 | 7.79 | 7.88 | 7.89 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 28 | 26 | 2 | 28 |
| DATE | | 3/6 | 3/7 | 3/8 | 3/9 | 3/10 | 3/6/07 | 3/7 | 3/8 | 3/8 | 3/9 | 3/10 | | | | | | | | | | | | | | | | | | |
| TIME | | 1600 | 1500 | 1535 | 1500 | 1410 | 1545 | 1015 | 1455 | 1459 | 1400 | 1400 | | | | | | | | | | | | | | | | | | |
| INITIALS | | SJ | MG | CS | CS | CS | SJ | CS | MT | CS | CS | CS | | | | | | | | | | | | | | | | | | |
| FED? | | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | | | | | | | | | | | | | | | | | | |

✖ - See:" EFFLUENT & DILUENT CHEMISTRY and WATER QUALITY DATA" sheet.
✦ -AERATE PRIOR TO MIXING DILUTIONS - AERATE TEST CHAMBERS FROM START!
◊ - "Old" water qualities (prior to renewal) ☆ - "New"water qualities (post renewal)

# ACUTE BIOASSAY DATA SUMMARY

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| STUDY: 15566 | SAMPLE RECEIVED: | "AS RECEIVED" EFFLUENT AND DILUENT CHEMISTRIES | | | | | |
| CLIENT: CH2M Hill | TEST ORGANISM: *A. bahia* | | TRC | AMM 0 HR✖ | AMM 48 HR✖ | pH | DO | Salinity |
| SAMPLE: American Samoa | ORGANISM SUPPLIER: | EFFLUENT | See "EFFLUENT & DILUENT CHEMISTRY and WATER QUALITY DATA" sheet | | | | | |
| DILUENT: LAB SALT | ORGANISM BATCH/AGE: | DILUENT | | | | | | |

| CONC | REP | SURVIVAL | | | | | ✦DISSOLVED OXYGEN (MG/L)✦ | | | | | | PH (SU) | | | | | | TEMPERATURE (°C) | | | | | | SALINITY (ppt) | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | 0 | 24 | 48 | 72 | 96 | 0 | 24 | 48◊ | 48☆ | 72 | 96 | 0 | 24 | 48◊ | 48☆ | 72 | 96 | 0 | 24 | 48◊ | 48 | 72 | 96 | 0 | 24 | 48◊ | 48 | 72 | 96 |
| 75% | A | 10 | 9 | 2 | — | — | 5.4 | 5.8 | — | 7.3 | 6.0 | 5.9 | 7.51 | 7.95 | 8.11 | 7.79 | 7.80 | — | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 28 | 26 | 27 | 27 |
| | B | 10 | 2 | 2 | 2 | — | 5.4 | 5.7 | 6.3 | 6.2 | 6.0 | 6.1 | 7.51 | 7.96 | 8.15 | 7.82 | 7.80 | 7.89 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 27 | 26 | 27 | 28 |
| | C | 10 | 5 | 2 | 2 | — | 5.4 | 5.9 | 5.9 | 6.2 | 6.0 | 6.1 | 7.50 | 7.97 | 8.19 | 7.81 | 7.80 | 7.81 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 28 | 26 | 26 | 28 |
| | D | 10 | 4 | 1 | — | — | 5.4 | 5.9 | 5.9 | 5.9 | 6.2 | 6.3 | 7.50 | 7.97 | 8.20 | 7.80 | 7.80 | 7.91 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 28 | 26 | 27 | 28 |
| | E | 10 | 0 | 0 | — | — | 5.4 | 5.8 | — | — | — | — | 7.50 | 7.96 | 8.22 | 7.82 | 7.80 | 7.91 | 20 | 20 | 19 | 20 | 19 | 20 | 25 | 26 | 28 | 26 | 27 | 28 |
| 100% | A | 10 | 1 | — | | | 5.0 | 4.5 | 5.1 | | | | 7.44 | 7.98 | 8.29 | | | | 20 | 20 | 19 | | | | 25 | 26 | 27 | | | |
| | B | 10 | 2 | — | | | 5.0 | 4.8 | 5.0 | | | | 7.43 | 7.97 | 8.25 | | | | 20 | 20 | 19 | | | | 25 | 26 | 28 | | | |
| | C | 10 | 0 | — | N/A | | 5.0 | 4.7 | — | | N/A | | 7.44 | 7.96 | | N/A | N/A | | 20 | 20 | 19 | | N/A | | 25 | 26 | 27 | N/A | N/A | |
| | D | 10 | 1 | — | | | 5.0 | 4.4 | 5.0 | | | | 7.44 | 7.93 | 8.27 | | | | 20 | 20 | 19 | | | | 25 | 26 | 28 | | | |
| | E | 10 | 0 | — | | | 5.0 | 4.8 | — | | | | 7.44 | 7.94 | | | | | 20 | 20 | 19 | | | | 25 | 26 | 27 | | | |
| DATE | | 3/6 | 3/7 | 3/8 | 3/9 | 3/10 | 3/6 | 3/7 | 3/8 | 3/8 | 3/9 | 3/10 | | | | | | | | | | | | | | | | | | |
| TIME | | 1600 | 1500 | 1945 | 1400 | 1410 | 1545 | 1550 | 1955 | 1350 | 1430 | 1400 | | | | | | | | | | | | | | | | | | |
| INITIALS | | SJ | MB | CS | CS | CS | SJ | MB | NT | CS | CS | CS | | | | | | | | | | | | | | | | | | |
| FED? | | ✓ | ✓ | ✓ | — | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | | | | | | | | | | | | | | | | | | |

✖ - See:" EFFLUENT & DILUENT CHEMISTRY and WATER QUALITY DATA" sheet.
✦ -AERATE PRIOR TO MIXING DILUTIONS - AERATE TEST CHAMBERS FROM START!
◊ - "Old" water qualities (prior to renewal) ☆ - "New"water qualities (post renewal)

# CETIS Test Summary

Report Date: 26 Mar-07 1:18 PM
Link: 08-8789-3478

**Americamysis 96-h Acute Survival Test** — EnviroSystems, Inc.

| | | | | | |
|---|---|---|---|---|---|
| Test No: | 18-5827-6829 | Test Type: | Survival (96h) | Duration: | 94h |
| Start Date: | 06 Mar-07 04:00 PM | Protocol: | EPA/821/R-02-012 (2002) | Species: | Americamysis bahia |
| Ending Date: | 10 Mar-07 02:00 PM | Dil Water: | Laboratory Seawater | Source: | ARO - Aquatic Research Organisms, N |
| Setup Date: | 06 Mar-07 04:00 PM | Brine: | Generic commercial salts | | |

| | | | | | |
|---|---|---|---|---|---|
| Sample No: | 01-9069-6116 | Material: | Industrial Effluent | Client: | CH2M Hill |
| Sample Date: | 28 Feb-07 12:00 PM | Code: | 15566 | Project: | First Quarter WET Compliance Test |
| Receive Date: | 06 Mar-07 09:30 AM | Source: | CH2M Hill- American Samoa | | |
| Sample Age: | 6d 4h (12 °C) | Station: | Joint Cannery Outfall | | |

**Comparison Summary**

| Analysis | Endpoint | NOEL | LOEL | ChV | MSDp | Method |
|---|---|---|---|---|---|---|
| 04-5943-0010 | 24h Proportion Survived | 50 | 75 | 61.237 | 16.68% | Dunnett's Multiple Comparison |
| 04-2705-1683 | 48h Proportion Survived | 25 | 50 | 35.355 | 15.05% | Dunnett's Multiple Comparison |
| 05-5196-3521 | 72h Proportion Survived | 25 | 50 | 35.355 | 17.52% | Dunnett's Multiple Comparison |
| 08-0745-3937 | 96h Proportion Survived | 25 | 50 | 35.355 | 19.94% | Dunnett's Multiple Comparison |

**Point Estimate Summary**

| Analysis | Endpoint | % Effect | Conc-% | 95% LCL | 95% UCL | Method |
|---|---|---|---|---|---|---|
| 05-8669-4874 | 24h Proportion Survived | 50 | 64.91535 | 59.83974 | 69.60133 | Linear Regression |
| 13-3332-7239 | 48h Proportion Survived | 50 | 57.21603 | 53.05392 | 61.06961 | Linear Regression |
| 02-7823-9229 | 72h Proportion Survived | 50 | 49.87932 | 42.70247 | 54.83716 | Linear Regression |
| 08-0185-1552 | 96h Proportion Survived | 50 | 44.05642 | 26.59244 | 58.86287 | Nonlinear Regression |

**Test Acceptability**

| Analysis | Endpoint | Attribute | Statistic | Acceptable Range | Decision |
|---|---|---|---|---|---|
| 08-0185-1552 | 96h Proportion Survived | Control Response | 0.96 | 0.9 - N/A | Passes acceptability criteria |
| 08-0745-3937 | 96h Proportion Survived | Control Response | 0.96 | 0.9 - N/A | Passes acceptability criteria |

# CETIS Test Summary

Report Date: 26 Mar-07 1:18 PM
Link: 08-8789-3478

**24h Proportion Survived Summary**

| Conc-% | Control Type | Reps | Mean | Minimum | Maximum | SE | SD | CV |
|---|---|---|---|---|---|---|---|---|
| 0 | Lab Water | 5 | 0.96000 | 0.90000 | 1.00000 | 0.02449 | 0.05477 | 5.71% |
| 6.25 | | 5 | 0.96000 | 0.90000 | 1.00000 | 0.02449 | 0.05477 | 5.71% |
| 12.5 | | 5 | 0.98000 | 0.90000 | 1.00000 | 0.02000 | 0.04472 | 4.56% |
| 25 | | 5 | 0.98000 | 0.90000 | 1.00000 | 0.02000 | 0.04472 | 4.56% |
| 50 | | 5 | 0.84000 | 0.70000 | 1.00000 | 0.06782 | 0.15166 | 18.05% |
| 75 | | 5 | 0.22000 | 0.00000 | 0.50000 | 0.10198 | 0.22804 | 103.65 |
| 100 | | 5 | 0.08000 | 0.00000 | 0.20000 | 0.03742 | 0.08367 | 104.58 |

**48h Proportion Survived Summary**

| Conc-% | Control Type | Reps | Mean | Minimum | Maximum | SE | SD | CV |
|---|---|---|---|---|---|---|---|---|
| 0 | Lab Water | 5 | 0.96000 | 0.90000 | 1.00000 | 0.02449 | 0.05477 | 5.71% |
| 6.25 | | 5 | 0.94000 | 0.90000 | 1.00000 | 0.02449 | 0.05477 | 5.83% |
| 12.5 | | 5 | 0.92000 | 0.70000 | 1.00000 | 0.05831 | 0.13038 | 14.17% |
| 25 | | 5 | 0.98000 | 0.90000 | 1.00000 | 0.02000 | 0.04472 | 4.56% |
| 50 | | 5 | 0.70000 | 0.60000 | 1.00000 | 0.07746 | 0.17321 | 24.74% |
| 75 | | 5 | 0.10000 | 0.00000 | 0.20000 | 0.04472 | 0.10000 | 100.00 |
| 100 | | 5 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00% |

**72h Proportion Survived Summary**

| Conc-% | Control Type | Reps | Mean | Minimum | Maximum | SE | SD | CV |
|---|---|---|---|---|---|---|---|---|
| 0 | Lab Water | 5 | 0.96000 | 0.90000 | 1.00000 | 0.02449 | 0.05477 | 5.71% |
| 6.25 | | 5 | 0.86000 | 0.80000 | 0.90000 | 0.02449 | 0.05477 | 6.37% |
| 12.5 | | 5 | 0.78000 | 0.60000 | 1.00000 | 0.09165 | 0.20494 | 26.27% |
| 25 | | 5 | 0.88000 | 0.80000 | 0.90000 | 0.02000 | 0.04472 | 5.08% |
| 50 | | 5 | 0.42000 | 0.10000 | 0.60000 | 0.08602 | 0.19235 | 45.80% |
| 75 | | 5 | 0.08000 | 0.00000 | 0.20000 | 0.04899 | 0.10954 | 136.93 |
| 100 | | 5 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00% |

**96h Proportion Survived Summary**

| Conc-% | Control Type | Reps | Mean | Minimum | Maximum | SE | SD | CV |
|---|---|---|---|---|---|---|---|---|
| 0 | Lab Water | 5 | 0.96000 | 0.90000 | 1.00000 | 0.02449 | 0.05477 | 5.71% |
| 6.25 | | 5 | 0.86000 | 0.80000 | 0.90000 | 0.02449 | 0.05477 | 6.37% |
| 12.5 | | 5 | 0.78000 | 0.60000 | 1.00000 | 0.09165 | 0.20494 | 26.27% |
| 25 | | 5 | 0.80000 | 0.50000 | 0.90000 | 0.07746 | 0.17321 | 21.65% |
| 50 | | 5 | 0.36000 | 0.10000 | 0.60000 | 0.09274 | 0.20736 | 57.60% |
| 75 | | 5 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00% |
| 100 | | 5 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00% |

# CETIS Test Summary

Report Date: 26 Mar-07 1:18 PM
Link: 08-8789-3478

**24h Proportion Survived Detail**

| Conc-% | Control Type | Rep 1 | Rep 2 | Rep 3 | Rep 4 | Rep 5 |
|---|---|---|---|---|---|---|
| 0 | Lab Water | 0.90000 | 1.00000 | 1.00000 | 0.90000 | 1.00000 |
| 6.25 | | 0.90000 | 1.00000 | 0.90000 | 1.00000 | 1.00000 |
| 12.5 | | 1.00000 | 1.00000 | 0.90000 | 1.00000 | 1.00000 |
| 25 | | 1.00000 | 1.00000 | 1.00000 | 1.00000 | 0.90000 |
| 50 | | 0.70000 | 1.00000 | 0.70000 | 1.00000 | 0.80000 |
| 75 | | 0.00000 | 0.20000 | 0.50000 | 0.40000 | 0.00000 |
| 100 | | 0.10000 | 0.20000 | 0.00000 | 0.10000 | 0.00000 |

**48h Proportion Survived Detail**

| Conc-% | Control Type | Rep 1 | Rep 2 | Rep 3 | Rep 4 | Rep 5 |
|---|---|---|---|---|---|---|
| 0 | Lab Water | 0.90000 | 1.00000 | 1.00000 | 0.90000 | 1.00000 |
| 6.25 | | 0.90000 | 1.00000 | 0.90000 | 1.00000 | 0.90000 |
| 12.5 | | 1.00000 | 1.00000 | 0.70000 | 1.00000 | 0.90000 |
| 25 | | 1.00000 | 1.00000 | 1.00000 | 1.00000 | 0.90000 |
| 50 | | 0.70000 | 0.60000 | 0.60000 | 1.00000 | 0.60000 |
| 75 | | 0.00000 | 0.20000 | 0.20000 | 0.10000 | 0.00000 |
| 100 | | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 |

**72h Proportion Survived Detail**

| Conc-% | Control Type | Rep 1 | Rep 2 | Rep 3 | Rep 4 | Rep 5 |
|---|---|---|---|---|---|---|
| 0 | Lab Water | 0.90000 | 1.00000 | 1.00000 | 0.90000 | 1.00000 |
| 6.25 | | 0.90000 | 0.90000 | 0.80000 | 0.90000 | 0.80000 |
| 12.5 | | 1.00000 | 1.00000 | 0.70000 | 0.60000 | 0.60000 |
| 25 | | 0.90000 | 0.80000 | 0.90000 | 0.90000 | 0.90000 |
| 50 | | 0.50000 | 0.40000 | 0.10000 | 0.60000 | 0.50000 |
| 75 | | 0.00000 | 0.20000 | 0.20000 | 0.00000 | 0.00000 |
| 100 | | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 |

**96h Proportion Survived Detail**

| Conc-% | Control Type | Rep 1 | Rep 2 | Rep 3 | Rep 4 | Rep 5 |
|---|---|---|---|---|---|---|
| 0 | Lab Water | 0.90000 | 1.00000 | 1.00000 | 0.90000 | 1.00000 |
| 6.25 | | 0.90000 | 0.90000 | 0.80000 | 0.90000 | 0.80000 |
| 12.5 | | 1.00000 | 1.00000 | 0.70000 | 0.60000 | 0.60000 |
| 25 | | 0.90000 | 0.80000 | 0.50000 | 0.90000 | 0.90000 |
| 50 | | 0.20000 | 0.40000 | 0.10000 | 0.60000 | 0.50000 |
| 75 | | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 |
| 100 | | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 |

# CETIS Analysis Detail

**Americamysis 96-h Acute Survival Test** — **EnviroSystems, Inc.**

| | | | | | |
|---|---|---|---|---|---|
| **Test No:** | 18-5827-6829 | **Test Type:** | Survival (96h) | **Duration:** | 94h |
| **Start Date:** | 06 Mar-07 04:00 PM | **Protocol:** | EPA/821/R-02-012 (2002) | **Species:** | Americamysis bahia |
| **Ending Date:** | 10 Mar-07 02:00 PM | **Dil Water:** | Laboratory Seawater | **Source:** | ARO - Aquatic Research Organisms, N |
| **Setup Date:** | 06 Mar-07 04:00 PM | **Brine:** | Generic commercial salts | | |

| | | | | | |
|---|---|---|---|---|---|
| **Sample No:** | 01-9069-6116 | **Material:** | Industrial Effluent | **Client:** | CH2M Hill |
| **Sample Date:** | 28 Feb-07 12:00 PM | **Code:** | 15566 | **Project:** | First Quarter WET Compliance Test |
| **Receive Date:** | 06 Mar-07 09:30 AM | **Source:** | CH2M Hill- American Samoa | | |
| **Sample Age:** | 6d 4h (12 °C) | **Station:** | Joint Cannery Outfall | | |

| Endpoint | Analysis Type | Sample Link | Control Link | Date Analyzed | Version |
|---|---|---|---|---|---|
| 24h Proportion Survived | Comparison | 08-8789-3478 | 08-8789-3478 | 13 Mar-07 8:48 AM | CETISv1.026 |

| Method | Alt H | Data Transform | Z | NOEL | LOEL | Toxic Units | ChV | MSDp |
|---|---|---|---|---|---|---|---|---|
| Dunnett's Multiple Comparison | C > T | Angular (Corrected) | | 50 | 75 | 2.00 | 61.237 | 16.68% |

**ANOVA Assumptions**

| Attribute | Test | Statistic | Critical | P Level | Decision(0.01) |
|---|---|---|---|---|---|
| Variances | Bartlett | 14.27727 | 16.81190 | 0.02669 | Equal Variances |
| Distribution | Shapiro-Wilk W | 0.94330 | 0.91004 | 0.09529 | Normal Distribution |

**ANOVA Table**

| Source | Sum of Squares | Mean Square | DF | F Statistic | P Level | Decision(0.05) |
|---|---|---|---|---|---|---|
| Between | 6.778985 | 1.129831 | 6 | 45.56 | 0.00000 | Significant Effect |
| Error | 0.6943628 | 0.0247987 | 28 | | | |
| Total | 7.47334731 | 1.1546294 | 34 | | | |

**Group Comparisons**

| Control | vs | Conc-% | Statistic | Critical | P Level | MSD | Decision(0.05) |
|---|---|---|---|---|---|---|---|
| Lab Water | | 6.25 | 0 | 2.40857 | > 0.0500 | 0.23989 | Non-Significant Effect |
| | | 12.5 | -0.3273 | 2.40857 | > 0.0500 | 0.23989 | Non-Significant Effect |
| | | 25 | -0.3273 | 2.40857 | > 0.0500 | 0.23989 | Non-Significant Effect |
| | | 50 | 1.64794 | 2.40857 | > 0.0500 | 0.23989 | Non-Significant Effect |
| | | 75 | 9.00195 | 2.40857 | <= 0.0500 | 0.23989 | Significant Effect |
| | | 100 | 10.6619 | 2.40857 | <= 0.0500 | 0.23989 | Significant Effect |

**Data Summary**

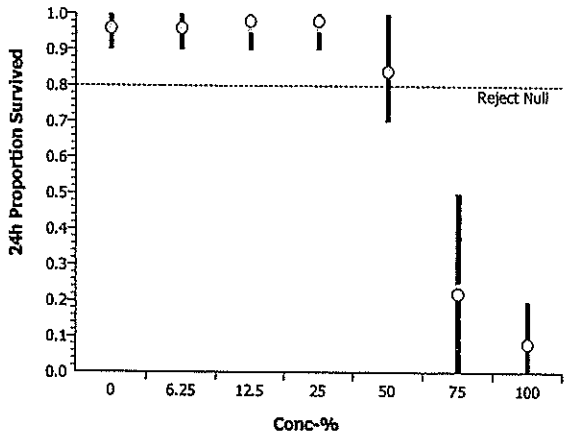
| | | | Original Data | | | | Transformed Data | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Conc-% | Control Type | Count | Mean | Minimum | Maximum | SD | Mean | Minimum | Maximum | SD |
| 0 | Lab Water | 5 | 0.96000 | 0.90000 | 1.00000 | 0.05477 | 1.34683 | 1.24905 | 1.41202 | 0.08926 |
| 6.25 | | 5 | 0.96000 | 0.90000 | 1.00000 | 0.05477 | 1.34683 | 1.24905 | 1.41202 | 0.08926 |
| 12.5 | | 5 | 0.98000 | 0.90000 | 1.00000 | 0.04472 | 1.37942 | 1.24905 | 1.41202 | 0.07288 |
| 25 | | 5 | 0.98000 | 0.90000 | 1.00000 | 0.04472 | 1.37942 | 1.24905 | 1.41202 | 0.07288 |
| 50 | | 5 | 0.84000 | 0.70000 | 1.00000 | 0.15166 | 1.18270 | 0.99116 | 1.41202 | 0.21463 |
| 75 | | 5 | 0.22000 | 0.00000 | 0.50000 | 0.22804 | 0.45027 | 0.15878 | 0.78540 | 0.29043 |
| 100 | | 5 | 0.08000 | 0.00000 | 0.20000 | 0.08367 | 0.28494 | 0.15878 | 0.46365 | 0.12892 |

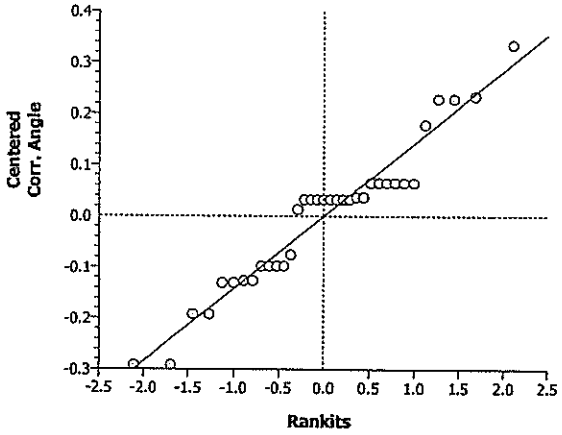
**Data Detail**

| Conc-% | Control Type | Rep 1 | Rep 2 | Rep 3 | Rep 4 | Rep 5 | Rep 6 | Rep 7 | Rep 8 | Rep 9 | Rep 10 |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 0 | Lab Water | 0.90000 | 1.00000 | 1.00000 | 0.90000 | 1.00000 | | | | | |
| 6.25 | | 0.90000 | 1.00000 | 0.90000 | 1.00000 | 1.00000 | | | | | |
| 12.5 | | 1.00000 | 1.00000 | 0.90000 | 1.00000 | 1.00000 | | | | | |
| 25 | | 1.00000 | 1.00000 | 1.00000 | 1.00000 | 0.90000 | | | | | |
| 50 | | 0.70000 | 1.00000 | 0.70000 | 1.00000 | 0.80000 | | | | | |
| 75 | | 0.00000 | 0.20000 | 0.50000 | 0.40000 | 0.00000 | | | | | |
| 100 | | 0.10000 | 0.20000 | 0.00000 | 0.10000 | 0.00000 | | | | | |

# CETIS Analysis Detail

**Graphics**



# CETIS Analysis Detail

**Americamysis 96-h Acute Survival Test** — **EnviroSystems, Inc.**

| | | | | | |
|---|---|---|---|---|---|
| **Test No:** | 18-5827-6829 | **Test Type:** | Survival (96h) | **Duration:** | 94h |
| **Start Date:** | 06 Mar-07 04:00 PM | **Protocol:** | EPA/821/R-02-012 (2002) | **Species:** | Americamysis bahia |
| **Ending Date:** | 10 Mar-07 02:00 PM | **Dil Water:** | Laboratory Seawater | **Source:** | ARO - Aquatic Research Organisms, N |
| **Setup Date:** | 06 Mar-07 04:00 PM | **Brine:** | Generic commercial salts | | |

| | | | | | |
|---|---|---|---|---|---|
| **Sample No:** | 01-9069-6116 | **Material:** | Industrial Effluent | **Client:** | CH2M Hill |
| **Sample Date:** | 28 Feb-07 12:00 PM | **Code:** | 15566 | **Project:** | First Quarter WET Compliance Test |
| **Receive Date:** | 06 Mar-07 09:30 AM | **Source:** | CH2M Hill- American Samoa | | |
| **Sample Age:** | 6d 4h (12 °C) | **Station:** | Joint Cannery Outfall | | |

| Endpoint | Analysis Type | Sample Link | Control Link | Date Analyzed | Version |
|---|---|---|---|---|---|
| 24h Proportion Survived | Linear Regression | 08-8789-3478 | 08-8789-3478 | 13 Mar-07 8:49 AM | CETISv1.026 |

**Linear Regression Options**

| Model | Threshold Option | Lower Threshold | Threshold Optimized | Reweighted | Pooled Groups | Heterogeneity Corr. |
|---|---|---|---|---|---|---|
| Log-Normal | Control Threshold | 0.04 | Yes | Yes | No | No |

**Regression Parameters**

| Parameter | Estimate | Std Error | 95% LCL | 95% UCL | t Statistic | P Level | Decision(0.05) |
|---|---|---|---|---|---|---|---|
| Threshold | 0.02932 | 0.01194 | 0.00592 | 0.05273 | 2.455 | 0.07004 | Not Significant |
| Slope | 8.64516 | 1.20893 | 6.27566 | 11.01466 | 7.151 | 0.00202 | Significant |
| Intercept | -10.66803 | 2.23053 | -15.03986 | -6.29620 | -4.783 | 0.00876 | Significant |

**Regression Summary**

| Iters | Log Likelihood | Mu | Sigma | G Stat | Chi-Sq | Critical | P Level | Decision(0.05) |
|---|---|---|---|---|---|---|---|---|
| 7 | -21.02848 | -1.23399 | 0.11567 | 0.07512 | 34.54569 | 41.33714 | 0.18348 | Non-Significant Heterogeneity |

**Residual Analysis**

| Attribute | Method | Statistic | Critical | P Level | Decision(0.05) |
|---|---|---|---|---|---|
| Variances | Modified Levene | 4.50173 | 2.52766 | 0.00372 | Unequal Variances |
| Distribution | Shapiro-Wilk W | 0.89931 | 0.92671 | 0.00971 | Non-normal Distribution |

**Point Estimates**

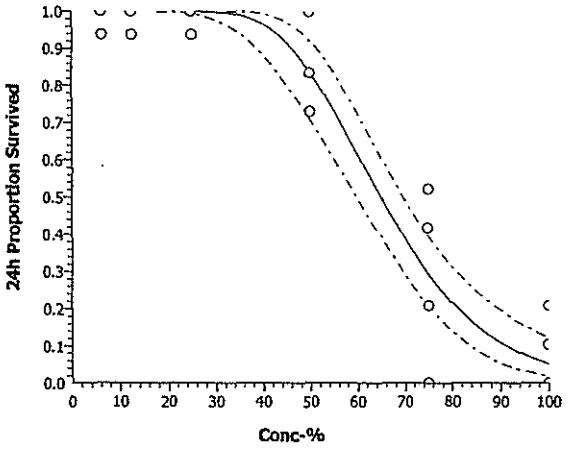
| % Effect | Conc-% | 95% LCL | 95% UCL |
|---|---|---|---|
| 50 | 64.91535 | 59.83974 | 69.60133 |

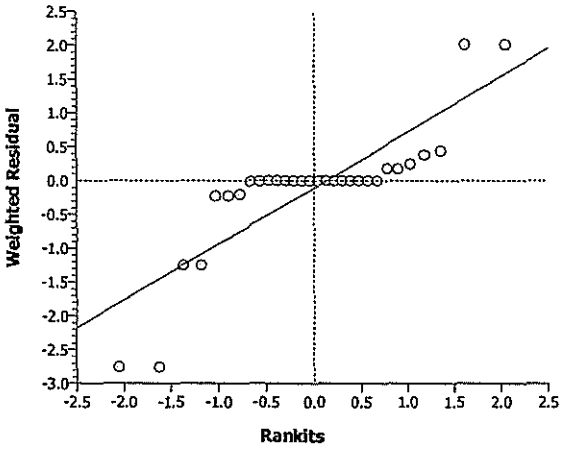
**Data Summary**

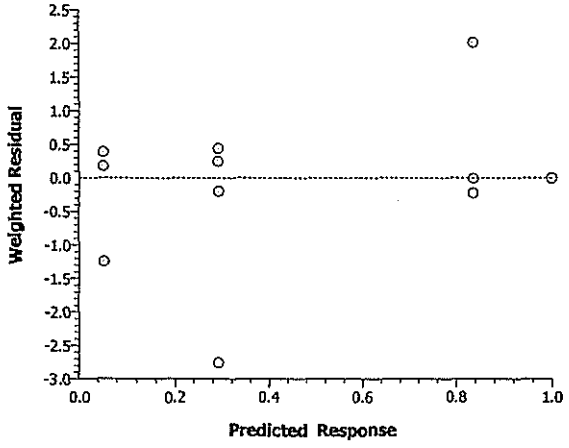
| | | | Calculated Variate(A/B) | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Conc-% | Control Type | Count | Mean | Minimum | Maximum | SE | SD | A | B |
| 0 | Lab Water | 5 | 0.96000 | 0.90000 | 1.00000 | 0.01118 | 0.05477 | 48 | 50 |
| 6.25 | | 5 | 0.96000 | 0.90000 | 1.00000 | 0.01118 | 0.05477 | 48 | 50 |
| 12.5 | | 5 | 0.98000 | 0.90000 | 1.00000 | 0.00913 | 0.04472 | 49 | 50 |
| 25 | | 5 | 0.98000 | 0.90000 | 1.00000 | 0.00913 | 0.04472 | 49 | 50 |
| 50 | | 5 | 0.84000 | 0.70000 | 1.00000 | 0.03096 | 0.15166 | 42 | 50 |
| 75 | | 5 | 0.22000 | 0.00000 | 0.50000 | 0.04655 | 0.22804 | 11 | 50 |
| 100 | | 5 | 0.08000 | 0.00000 | 0.20000 | 0.01708 | 0.08367 | 4 | 50 |

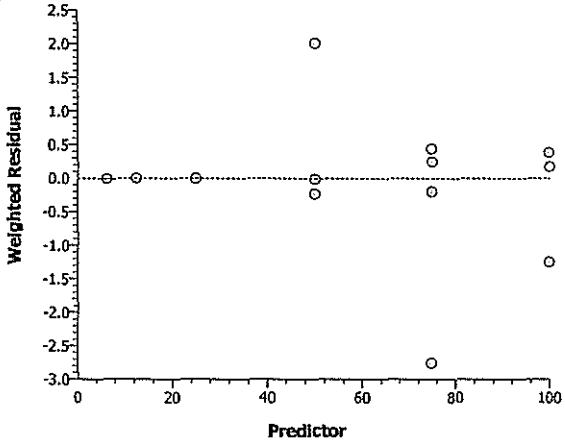
# CETIS Analysis Detail

**Graphics**



# CETIS Analysis Detail

**Americamysis 96-h Acute Survival Test** — **EnviroSystems, Inc.**

| | | | | | |
|---|---|---|---|---|---|
| **Test No:** | 18-5827-6829 | **Test Type:** | Survival (96h) | **Duration:** | 94h |
| **Start Date:** | 06 Mar-07 04:00 PM | **Protocol:** | EPA/821/R-02-012 (2002) | **Species:** | Americamysis bahia |
| **Ending Date:** | 10 Mar-07 02:00 PM | **Dil Water:** | Laboratory Seawater | **Source:** | ARO - Aquatic Research Organisms, N |
| **Setup Date:** | 06 Mar-07 04:00 PM | **Brine:** | Generic commercial salts | | |

| | | | | | |
|---|---|---|---|---|---|
| **Sample No:** | 01-9069-6116 | **Material:** | Industrial Effluent | **Client:** | CH2M Hill |
| **Sample Date:** | 28 Feb-07 12:00 PM | **Code:** | 15566 | **Project:** | First Quarter WET Compliance Test |
| **Receive Date:** | 06 Mar-07 09:30 AM | **Source:** | CH2M Hill- American Samoa | | |
| **Sample Age:** | 6d 4h (12 °C) | **Station:** | Joint Cannery Outfall | | |

| Endpoint | Analysis Type | Sample Link | Control Link | Date Analyzed | Version |
|---|---|---|---|---|---|
| 48h Proportion Survived | Comparison | 08-8789-3478 | 08-8789-3478 | 13 Mar-07 8:48 AM | CETISv1.026 |

| Method | Alt H | Data Transform | Z | NOEL | LOEL | Toxic Units | ChV | MSDp |
|---|---|---|---|---|---|---|---|---|
| Dunnett's Multiple Comparison | C > T | Angular (Corrected) | | 25 | 50 | 4.00 | 35.355 | 15.05% |

**ANOVA Assumptions**

| Attribute | Test | Statistic | Critical | P Level | Decision(0.01) |
|---|---|---|---|---|---|
| Variances | Bartlett | 7.40419 | 15.08628 | 0.19227 | Equal Variances |
| Distribution | Shapiro-Wilk W | 0.95435 | 0.89981 | 0.25762 | Normal Distribution |

**ANOVA Table**

| Source | Sum of Squares | Mean Square | DF | F Statistic | P Level | Decision(0.05) |
|---|---|---|---|---|---|---|
| Between | 4.244753 | 0.8489506 | 5 | 39.07 | 0.00000 | Significant Effect |
| Error | 0.5215501 | 0.0217313 | 24 | | | |
| Total | 4.76630294 | 0.8706819 | 29 | | | |

**Group Comparisons**

| Control | vs | Conc-% | Statistic | Critical | P Level | MSD | Decision(0.05) |
|---|---|---|---|---|---|---|---|
| Lab Water | | 6.25 | 0.3496 | 2.36 | > 0.0500 | 0.22003 | Non-Significant Effect |
| | | 12.5 | 0.55321 | 2.36 | > 0.0500 | 0.22003 | Non-Significant Effect |
| | | 25 | -0.3496 | 2.36 | > 0.0500 | 0.22003 | Non-Significant Effect |
| | | 50 | 3.58827 | 2.36 | <= 0.0500 | 0.22003 | Significant Effect |
| | | 75 | 11.0851 | 2.36 | <= 0.0500 | 0.22003 | Significant Effect |

**Data Summary**

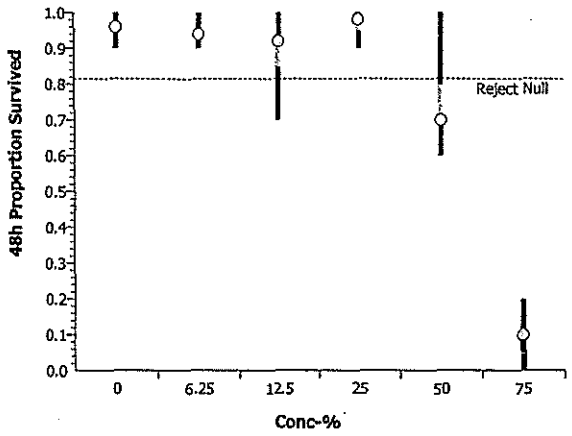
| | | | Original Data | | | | Transformed Data | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Conc-% | Control Type | Count | Mean | Minimum | Maximum | SD | Mean | Minimum | Maximum | SD |
| 0 | Lab Water | 5 | 0.96000 | 0.90000 | 1.00000 | 0.05477 | 1.34683 | 1.24905 | 1.41202 | 0.08926 |
| 6.25 | | 5 | 0.94000 | 0.90000 | 1.00000 | 0.05477 | 1.31423 | 1.24905 | 1.41202 | 0.08926 |
| 12.5 | | 5 | 0.92000 | 0.70000 | 1.00000 | 0.13038 | 1.29525 | 0.99116 | 1.41202 | 0.18406 |
| 25 | | 5 | 0.98000 | 0.90000 | 1.00000 | 0.04472 | 1.37942 | 1.24905 | 1.41202 | 0.07288 |
| 50 | | 5 | 0.70000 | 0.60000 | 1.00000 | 0.17321 | 1.01228 | 0.88608 | 1.41202 | 0.22804 |
| 75 | | 5 | 0.10000 | 0.00000 | 0.20000 | 0.10000 | 0.31332 | 0.15878 | 0.46365 | 0.15251 |

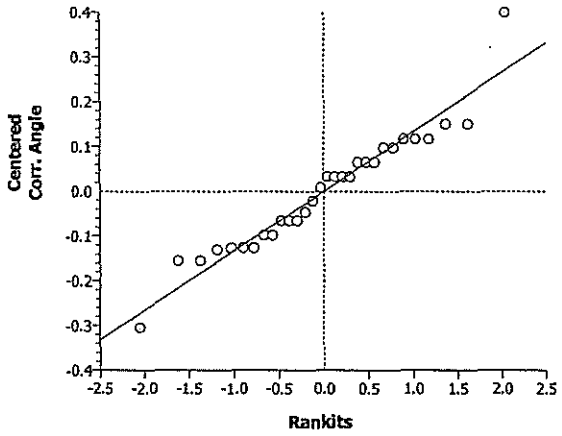
**Data Detail**

| Conc-% | Control Type | Rep 1 | Rep 2 | Rep 3 | Rep 4 | Rep 5 | Rep 6 | Rep 7 | Rep 8 | Rep 9 | Rep 10 |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 0 | Lab Water | 0.90000 | 1.00000 | 1.00000 | 0.90000 | 1.00000 | | | | | |
| 6.25 | | 0.90000 | 1.00000 | 0.90000 | 1.00000 | 0.90000 | | | | | |
| 12.5 | | 1.00000 | 1.00000 | 0.70000 | 1.00000 | 0.90000 | | | | | |
| 25 | | 1.00000 | 1.00000 | 1.00000 | 1.00000 | 0.90000 | | | | | |
| 50 | | 0.70000 | 0.60000 | 0.60000 | 1.00000 | 0.60000 | | | | | |
| 75 | | 0.00000 | 0.20000 | 0.20000 | 0.10000 | 0.00000 | | | | | |

# CETIS Analysis Detail

**Graphics**



# CETIS Analysis Detail

**Americamysis 96-h Acute Survival Test** — **EnviroSystems, Inc.**

| | | | | | |
|---|---|---|---|---|---|
| **Test No:** | 18-5827-6829 | **Test Type:** | Survival (96h) | **Duration:** | 94h |
| **Start Date:** | 06 Mar-07 04:00 PM | **Protocol:** | EPA/821/R-02-012 (2002) | **Species:** | Americamysis bahia |
| **Ending Date:** | 10 Mar-07 02:00 PM | **Dil Water:** | Laboratory Seawater | **Source:** | ARO - Aquatic Research Organisms, N |
| **Setup Date:** | 06 Mar-07 04:00 PM | **Brine:** | Generic commercial salts | | |

| | | | | | |
|---|---|---|---|---|---|
| **Sample No:** | 01-9069-6116 | **Material:** | Industrial Effluent | **Client:** | CH2M Hill |
| **Sample Date:** | 28 Feb-07 12:00 PM | **Code:** | 15566 | **Project:** | First Quarter WET Compliance Test |
| **Receive Date:** | 06 Mar-07 09:30 AM | **Source:** | CH2M Hill- American Samoa | | |
| **Sample Age:** | 6d 4h (12 °C) | **Station:** | Joint Cannery Outfall | | |

| Endpoint | Analysis Type | Sample Link | Control Link | Date Analyzed | Version |
|---|---|---|---|---|---|
| 48h Proportion Survived | Linear Regression | 08-8789-3478 | 08-8789-3478 | 13 Mar-07 8:49 AM | CETISv1.026 |

**Linear Regression Options**

| Model | Threshold Option | Lower Threshold | Threshold Optimized | Reweighted | Pooled Groups | Heterogeneity Corr. |
|---|---|---|---|---|---|---|
| Log-Normal | Control Threshold | 0.04 | Yes | Yes | No | No |

**Regression Parameters**

| Parameter | Estimate | Std Error | 95% LCL | 95% UCL | t Statistic | P Level | Decision(0.05) |
|---|---|---|---|---|---|---|---|
| Threshold | 0.05013 | 0.01543 | 0.01988 | 0.08037 | 3.249 | 0.03142 | Significant |
| Slope | 11.10595 | 1.66268 | 7.84711 | 14.36480 | 6.680 | 0.00261 | Significant |
| Intercept | -14.51891 | 2.96394 | -20.32824 | -8.70958 | -4.899 | 0.00805 | Significant |

**Regression Summary**

| Iters | Log Likelihood | Mu | Sigma | G Stat | Chi-Sq | Critical | P Level | Decision(0.05) |
|---|---|---|---|---|---|---|---|---|
| 8 | -60.23325 | -1.30731 | 0.09004 | 0.08610 | 31.29955 | 41.33714 | 0.30394 | Non-Significant Heterogeneity |

**Residual Analysis**

| Attribute | Method | Statistic | Critical | P Level | Decision(0.05) |
|---|---|---|---|---|---|
| Variances | Modified Levene | 2.30535 | 2.52766 | 0.06876 | Equal Variances |
| Distribution | Shapiro-Wilk W | 0.75249 | 0.92671 | 0.00000 | Non-normal Distribution |

**Point Estimates**

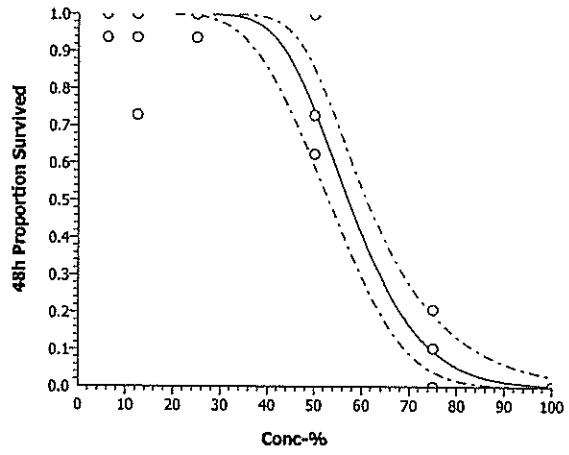
| % Effect | Conc-% | 95% LCL | 95% UCL |
|---|---|---|---|
| 50 | 57.21603 | 53.05392 | 61.06961 |

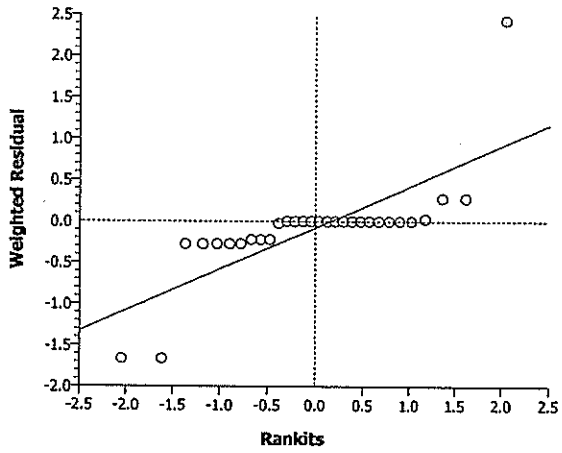
**Data Summary**

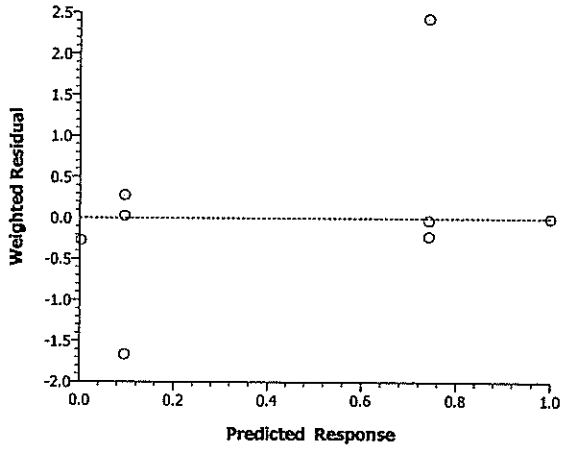
| | | | Calculated Variate(A/B) | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Conc-% | Control Type | Count | Mean | Minimum | Maximum | SE | SD | A | B |
| 0 | Lab Water | 5 | 0.96000 | 0.90000 | 1.00000 | 0.01118 | 0.05477 | 48 | 50 |
| 6.25 | | 5 | 0.94000 | 0.90000 | 1.00000 | 0.01118 | 0.05477 | 47 | 50 |
| 12.5 | | 5 | 0.92000 | 0.70000 | 1.00000 | 0.02661 | 0.13038 | 46 | 50 |
| 25 | | 5 | 0.98000 | 0.90000 | 1.00000 | 0.00913 | 0.04472 | 49 | 50 |
| 50 | | 5 | 0.70000 | 0.60000 | 1.00000 | 0.03536 | 0.17321 | 35 | 50 |
| 75 | | 5 | 0.10000 | 0.00000 | 0.20000 | 0.02041 | 0.10000 | 5 | 50 |
| 100 | | 5 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0 | 50 |

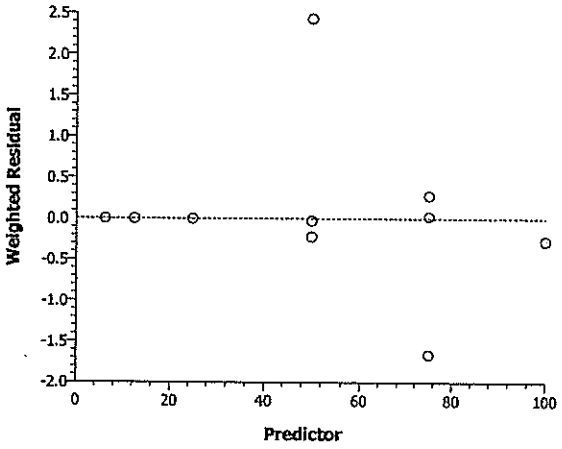
# CETIS Analysis Detail

**Graphics**



# CETIS Analysis Detail

**Americamysis 96-h Acute Survival Test** — **EnviroSystems, Inc.**

| | | | | | |
|---|---|---|---|---|---|
| **Test No:** | 18-5827-6829 | **Test Type:** | Survival (96h) | **Duration:** | 94h |
| **Start Date:** | 06 Mar-07 04:00 PM | **Protocol:** | EPA/821/R-02-012 (2002) | **Species:** | Americamysis bahia |
| **Ending Date:** | 10 Mar-07 02:00 PM | **Dil Water:** | Laboratory Seawater | **Source:** | ARO - Aquatic Research Organisms, N |
| **Setup Date:** | 06 Mar-07 04:00 PM | **Brine:** | Generic commercial salts | | |

| | | | | | |
|---|---|---|---|---|---|
| **Sample No:** | 01-9069-6116 | **Material:** | Industrial Effluent | **Client:** | CH2M Hill |
| **Sample Date:** | 28 Feb-07 12:00 PM | **Code:** | 15566 | **Project:** | First Quarter WET Compliance Test |
| **Receive Date:** | 06 Mar-07 09:30 AM | **Source:** | CH2M Hill- American Samoa | | |
| **Sample Age:** | 6d 4h (12 °C) | **Station:** | Joint Cannery Outfall | | |

| Endpoint | Analysis Type | Sample Link | Control Link | Date Analyzed | Version |
|---|---|---|---|---|---|
| 72h Proportion Survived | Comparison | 08-8789-3478 | 08-8789-3478 | 13 Mar-07 8:48 AM | CETISv1.026 |

| Method | Alt H | Data Transform | Z | NOEL | LOEL | Toxic Units | ChV | MSDp |
|---|---|---|---|---|---|---|---|---|
| Dunnett's Multiple Comparison | C > T | Angular (Corrected) | | 25 | 50 | 4.00 | 35.355 | 17.52% |

**ANOVA Assumptions**

| Attribute | Test | Statistic | Critical | P Level | Decision(0.01) |
|---|---|---|---|---|---|
| Variances | Bartlett | 11.75380 | 15.08628 | 0.03832 | Equal Variances |
| Distribution | Shapiro-Wilk W | 0.96380 | 0.89981 | 0.43068 | Normal Distribution |

**ANOVA Table**

| Source | Sum of Squares | Mean Square | DF | F Statistic | P Level | Decision(0.05) |
|---|---|---|---|---|---|---|
| Between | 4.139223 | 0.8278446 | 5 | 29.54 | 0.00000 | Significant Effect |
| Error | 0.6724767 | 0.0280199 | 24 | | | |
| Total | 4.81169981 | 0.8558645 | 29 | | | |

**Group Comparisons**

| Control | vs | Conc-% | Statistic | Critical | P Level | MSD | Decision(0.05) |
|---|---|---|---|---|---|---|---|
| Lab Water | | 6.25 | 1.45976 | 2.36 | > 0.0500 | 0.24985 | Non-Significant Effect |
| | | 12.5 | 2.16647 | 2.36 | > 0.0500 | 0.24985 | Non-Significant Effect |
| | | 25 | 1.19169 | 2.36 | > 0.0500 | 0.24985 | Non-Significant Effect |
| | | 50 | 6.17903 | 2.36 | <= 0.0500 | 0.24985 | Significant Effect |
| | | 75 | 10.0701 | 2.36 | <= 0.0500 | 0.24985 | Significant Effect |

**Data Summary**

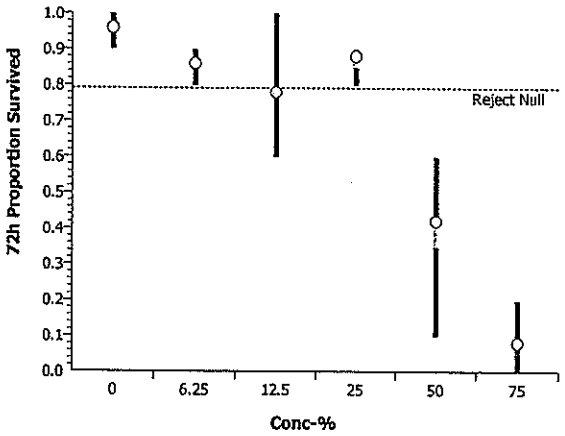
| | | | Original Data | | | | Transformed Data | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Conc-% | Control Type | Count | Mean | Minimum | Maximum | SD | Mean | Minimum | Maximum | SD |
| 0 | Lab Water | 5 | 0.96000 | 0.90000 | 1.00000 | 0.05477 | 1.34683 | 1.24905 | 1.41202 | 0.08926 |
| 6.25 | | 5 | 0.86000 | 0.80000 | 0.90000 | 0.05477 | 1.19229 | 1.10715 | 1.24905 | 0.07772 |
| 12.5 | | 5 | 0.78000 | 0.60000 | 1.00000 | 0.20494 | 1.11747 | 0.88608 | 1.41202 | 0.27228 |
| 25 | | 5 | 0.88000 | 0.80000 | 0.90000 | 0.04472 | 1.22067 | 1.10715 | 1.24905 | 0.06346 |
| 50 | | 5 | 0.42000 | 0.10000 | 0.60000 | 0.19235 | 0.69267 | 0.32175 | 0.88608 | 0.21923 |
| 75 | | 5 | 0.08000 | 0.00000 | 0.20000 | 0.10954 | 0.28073 | 0.15878 | 0.46365 | 0.16698 |

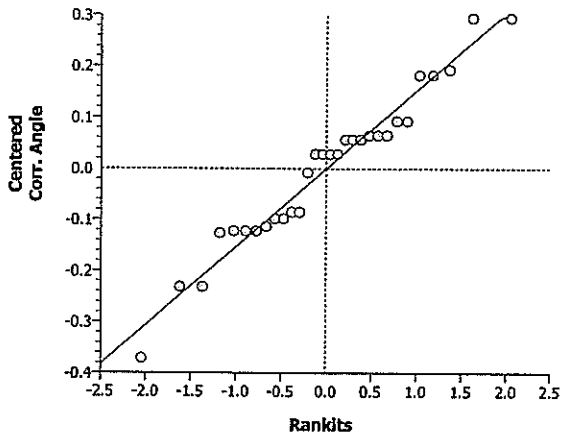
**Data Detail**

| Conc-% | Control Type | Rep 1 | Rep 2 | Rep 3 | Rep 4 | Rep 5 | Rep 6 | Rep 7 | Rep 8 | Rep 9 | Rep 10 |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 0 | Lab Water | 0.90000 | 1.00000 | 1.00000 | 0.90000 | 1.00000 | | | | | |
| 6.25 | | 0.90000 | 0.90000 | 0.80000 | 0.90000 | 0.80000 | | | | | |
| 12.5 | | 1.00000 | 1.00000 | 0.70000 | 0.60000 | 0.60000 | | | | | |
| 25 | | 0.90000 | 0.80000 | 0.90000 | 0.90000 | 0.90000 | | | | | |
| 50 | | 0.50000 | 0.40000 | 0.10000 | 0.60000 | 0.50000 | | | | | |
| 75 | | 0.00000 | 0.20000 | 0.20000 | 0.00000 | 0.00000 | | | | | |

# CETIS Analysis Detail

**Graphics**



# CETIS Analysis Detail

**Americamysis 96-h Acute Survival Test** — **EnviroSystems, Inc.**

| | | | | | |
|---|---|---|---|---|---|
| **Test No:** | 18-5827-6829 | **Test Type:** | Survival (96h) | **Duration:** | 94h |
| **Start Date:** | 06 Mar-07 04:00 PM | **Protocol:** | EPA/821/R-02-012 (2002) | **Species:** | Americamysis bahia |
| **Ending Date:** | 10 Mar-07 02:00 PM | **Dil Water:** | Laboratory Seawater | **Source:** | ARO - Aquatic Research Organisms, N |
| **Setup Date:** | 06 Mar-07 04:00 PM | **Brine:** | Generic commercial salts | | |

| | | | | | |
|---|---|---|---|---|---|
| **Sample No:** | 01-9069-6116 | **Material:** | Industrial Effluent | **Client:** | CH2M Hill |
| **Sample Date:** | 28 Feb-07 12:00 PM | **Code:** | 15566 | **Project:** | First Quarter WET Compliance Test |
| **Receive Date:** | 06 Mar-07 09:30 AM | **Source:** | CH2M Hill- American Samoa | | |
| **Sample Age:** | 6d 4h (12 °C) | **Station:** | Joint Cannery Outfall | | |

| Endpoint | Analysis Type | Sample Link | Control Link | Date Analyzed | Version |
|---|---|---|---|---|---|
| 72h Proportion Survived | Linear Regression | 08-8789-3478 | 08-8789-3478 | 13 Mar-07 8:49 AM | CETISv1.026 |

**Linear Regression Options**

| Model | Threshold Option | Lower Threshold | Threshold Optimized | Reweighted | Pooled Groups | Heterogeneity Corr. |
|---|---|---|---|---|---|---|
| Log-Normal | Control Threshold | 0.04 | Yes | Yes | No | No |

**Regression Parameters**

| Parameter | Estimate | Std Error | 95% LCL | 95% UCL | t Statistic | P Level | Decision(0.05) |
|---|---|---|---|---|---|---|---|
| Threshold | 0.12899 | 0.02424 | 0.08147 | 0.17651 | 5.320 | 0.00600 | Significant |
| Slope | 8.17900 | 1.60281 | 5.03749 | 11.32051 | 5.103 | 0.00697 | Significant |
| Intercept | -8.88729 | 2.84291 | -14.45940 | -3.31518 | -3.126 | 0.03532 | Significant |

**Regression Summary**

| Iters | Log Likelihood | Mu | Sigma | G Stat | Chi-Sq | Critical | P Level | Decision(0.05) |
|---|---|---|---|---|---|---|---|---|
| 9 | -114.52330 | -1.08660 | 0.12226 | 0.14753 | 35.10026 | 41.33714 | 0.16697 | Non-Significant Heterogeneity |

**Residual Analysis**

| Attribute | Method | Statistic | Critical | P Level | Decision(0.05) |
|---|---|---|---|---|---|
| Variances | Modified Levene | 2.90423 | 2.52766 | 0.02946 | Unequal Variances |
| Distribution | Shapiro-Wilk W | 0.71367 | 0.92671 | 0.00000 | Non-normal Distribution |

**Point Estimates**

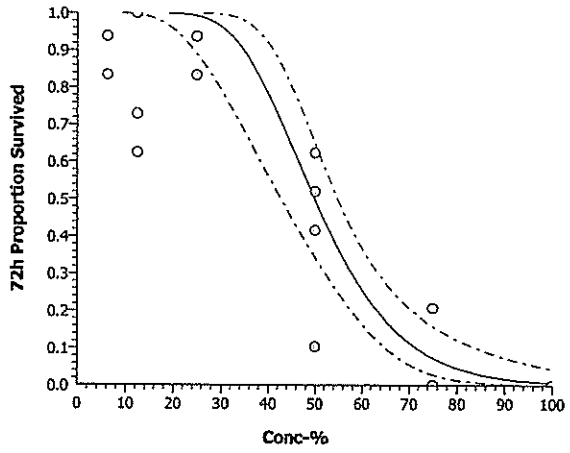
| % Effect | Conc-% | 95% LCL | 95% UCL |
|---|---|---|---|
| 50 | 49.87932 | 42.70247 | 54.83716 |

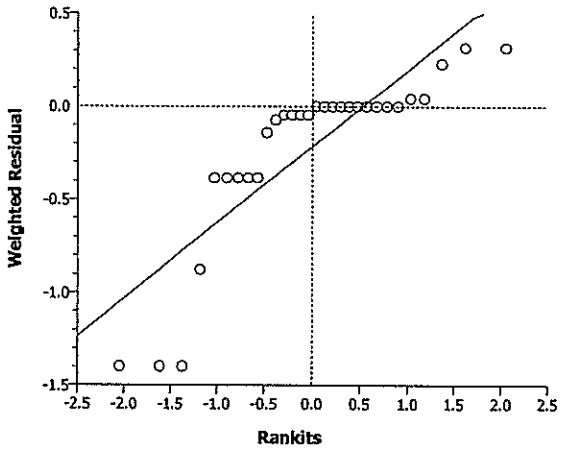
**Data Summary**

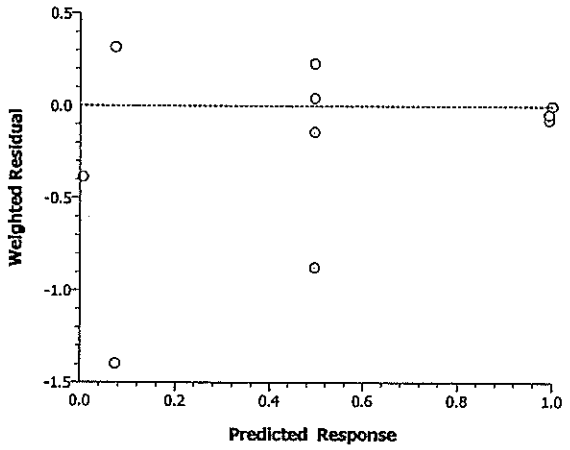
| | | | Calculated Variate(A/B) | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Conc-% | Control Type | Count | Mean | Minimum | Maximum | SE | SD | A | B |
| 0 | Lab Water | 5 | 0.96000 | 0.90000 | 1.00000 | 0.01118 | 0.05477 | 48 | 50 |
| 6.25 | | 5 | 0.86000 | 0.80000 | 0.90000 | 0.01118 | 0.05477 | 43 | 50 |
| 12.5 | | 5 | 0.78000 | 0.60000 | 1.00000 | 0.04183 | 0.20494 | 39 | 50 |
| 25 | | 5 | 0.88000 | 0.80000 | 0.90000 | 0.00913 | 0.04472 | 44 | 50 |
| 50 | | 5 | 0.42000 | 0.10000 | 0.60000 | 0.03926 | 0.19235 | 21 | 50 |
| 75 | | 5 | 0.08000 | 0.00000 | 0.20000 | 0.02236 | 0.10954 | 4 | 50 |
| 100 | | 5 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0 | 50 |

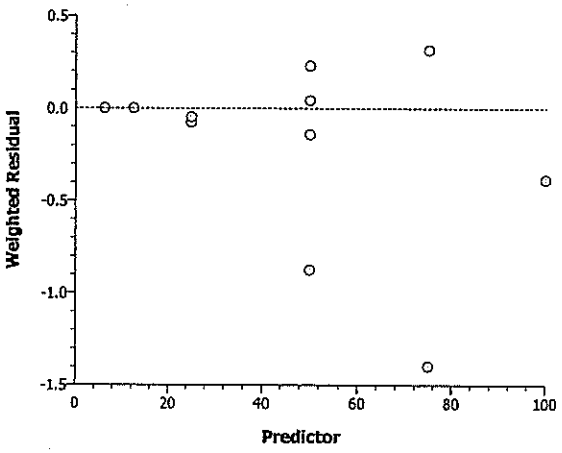
# CETIS Analysis Detail

**Graphics**



# CETIS Analysis Detail

**Americamysis 96-h Acute Survival Test** — **EnviroSystems, Inc.**

| | | | | | |
|---|---|---|---|---|---|
| **Test No:** | 18-5827-6829 | **Test Type:** | Survival (96h) | **Duration:** | 94h |
| **Start Date:** | 06 Mar-07 04:00 PM | **Protocol:** | EPA/821/R-02-012 (2002) | **Species:** | Americamysis bahia |
| **Ending Date:** | 10 Mar-07 02:00 PM | **Dil Water:** | Laboratory Seawater | **Source:** | ARO - Aquatic Research Organisms, N |
| **Setup Date:** | 06 Mar-07 04:00 PM | **Brine:** | Generic commercial salts | | |

| | | | | | |
|---|---|---|---|---|---|
| **Sample No:** | 01-9069-6116 | **Material:** | Industrial Effluent | **Client:** | CH2M Hill |
| **Sample Date:** | 28 Feb-07 12:00 PM | **Code:** | 15566 | **Project:** | First Quarter WET Compliance Test |
| **Receive Date:** | 06 Mar-07 09:30 AM | **Source:** | CH2M Hill- American Samoa | | |
| **Sample Age:** | 6d 4h (12 °C) | **Station:** | Joint Cannery Outfall | | |

| Endpoint | Analysis Type | Sample Link | Control Link | Date Analyzed | Version |
|---|---|---|---|---|---|
| 96h Proportion Survived | Comparison | 08-8789-3478 | 08-8789-3478 | 13 Mar-07 8:48 AM | CETISv1.026 |

| Method | Alt H | Data Transform | Z | NOEL | LOEL | Toxic Units | ChV | MSDp |
|---|---|---|---|---|---|---|---|---|
| Dunnett's Multiple Comparison | C > T | Angular (Corrected) | | 25 | 50 | 4.00 | 35.355 | 19.94% |

**Test Acceptability**

| Attribute | Statistic | Acceptable Range | Decision |
|---|---|---|---|
| Control Response | 0.96 | 0.9 - N/A | Passes acceptability criteria |

**ANOVA Assumptions**

| Attribute | Test | Statistic | Critical | P Level | Decision(0.01) |
|---|---|---|---|---|---|
| Variances | Bartlett | 7.70540 | 13.27671 | 0.10299 | Equal Variances |
| Distribution | Shapiro-Wilk W | 0.95720 | 0.88746 | 0.37074 | Normal Distribution |

**ANOVA Table**

| Source | Sum of Squares | Mean Square | DF | F Statistic | P Level | Decision(0.05) |
|---|---|---|---|---|---|---|
| Between | 1.457453 | 0.3643632 | 4 | 9.99 | 0.00013 | Significant Effect |
| Error | 0.7297502 | 0.0364875 | 20 | | | |
| Total | 2.18720293 | 0.4008507 | 24 | | | |

**Group Comparisons**

| Control | vs | Conc-% | Statistic | Critical | P Level | MSD | Decision(0.05) |
|---|---|---|---|---|---|---|---|
| Lab Water | | 6.25 | 1.27921 | 2.3 | > 0.0500 | 0.27786 | Non-Significant Effect |
| | | 12.5 | 1.89852 | 2.3 | > 0.0500 | 0.27786 | Non-Significant Effect |
| | | 25 | 1.81187 | 2.3 | > 0.0500 | 0.27786 | Non-Significant Effect |
| | | 50 | 5.94744 | 2.3 | <= 0.0500 | 0.27786 | Significant Effect |

**Data Summary**

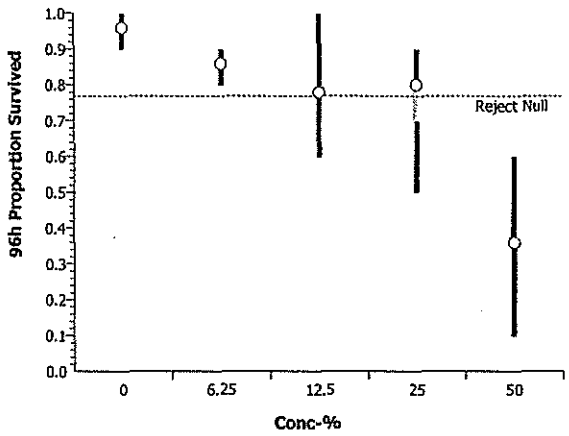
| | | | Original Data | | | | Transformed Data | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Conc-% | Control Type | Count | Mean | Minimum | Maximum | SD | Mean | Minimum | Maximum | SD |
| 0 | Lab Water | 5 | 0.96000 | 0.90000 | 1.00000 | 0.05477 | 1.34683 | 1.24905 | 1.41202 | 0.08926 |
| 6.25 | | 5 | 0.86000 | 0.80000 | 0.90000 | 0.05477 | 1.19229 | 1.10715 | 1.24905 | 0.07772 |
| 12.5 | | 5 | 0.78000 | 0.60000 | 1.00000 | 0.20494 | 1.11747 | 0.88608 | 1.41202 | 0.27228 |
| 25 | | 5 | 0.80000 | 0.50000 | 0.90000 | 0.17321 | 1.12794 | 0.78540 | 1.24905 | 0.20110 |
| 50 | | 5 | 0.36000 | 0.10000 | 0.60000 | 0.20736 | 0.62832 | 0.32175 | 0.88608 | 0.23205 |

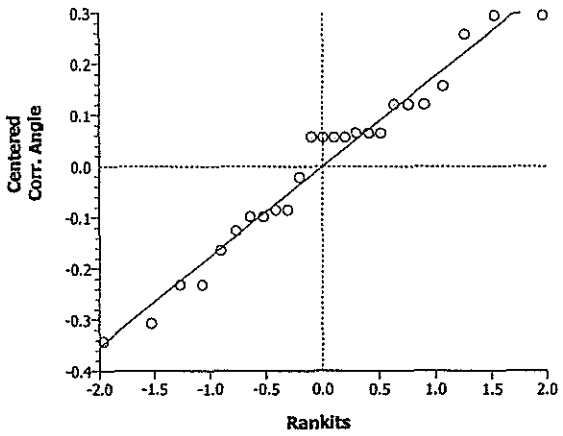
**Data Detail**

| Conc-% | Control Type | Rep 1 | Rep 2 | Rep 3 | Rep 4 | Rep 5 | Rep 6 | Rep 7 | Rep 8 | Rep 9 | Rep 10 |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 0 | Lab Water | 0.90000 | 1.00000 | 1.00000 | 0.90000 | 1.00000 | | | | | |
| 6.25 | | 0.90000 | 0.90000 | 0.80000 | 0.90000 | 0.80000 | | | | | |
| 12.5 | | 1.00000 | 1.00000 | 0.70000 | 0.60000 | 0.60000 | | | | | |
| 25 | | 0.90000 | 0.80000 | 0.50000 | 0.90000 | 0.90000 | | | | | |
| 50 | | 0.20000 | 0.40000 | 0.10000 | 0.60000 | 0.50000 | | | | | |

# CETIS Analysis Detail

**Graphics**



# CETIS Analysis Detail

**Americamysis 96-h Acute Survival Test** — **EnviroSystems, Inc.**

| | | | | | |
|---|---|---|---|---|---|
| **Test No:** | 18-5827-6829 | **Test Type:** | Survival (96h) | **Duration:** | 94h |
| **Start Date:** | 06 Mar-07 04:00 PM | **Protocol:** | EPA/821/R-02-012 (2002) | **Species:** | Americamysis bahia |
| **Ending Date:** | 10 Mar-07 02:00 PM | **Dil Water:** | Laboratory Seawater | **Source:** | ARO - Aquatic Research Organisms, N |
| **Setup Date:** | 06 Mar-07 04:00 PM | **Brine:** | Generic commercial salts | | |

| | | | | | |
|---|---|---|---|---|---|
| **Sample No:** | 01-9069-6116 | **Material:** | Industrial Effluent | **Client:** | CH2M Hill |
| **Sample Date:** | 28 Feb-07 12:00 PM | **Code:** | 15566 | **Project:** | First Quarter WET Compliance Test |
| **Receive Date:** | 06 Mar-07 09:30 AM | **Source:** | CH2M Hill- American Samoa | | |
| **Sample Age:** | 6d 4h (12 °C) | **Station:** | Joint Cannery Outfall | | |

| Endpoint | Analysis Type | Sample Link | Control Link | Date Analyzed | Version |
|---|---|---|---|---|---|
| 96h Proportion Survived | Nonlinear Regression | 08-8789-3478 | 08-8789-3478 | 13 Mar-07 8:49 AM | CETISv1.026 |

**Non-Linear Regression Options**

| X Transform | Y Transform | Model Function | Weighting Function | PTBS Function |
|---|---|---|---|---|
| None | None | 3 Parameter Logistic [Y=A/(1+Exp(-C(X-D)))] | No Weight [W = 1] | No Transform [None] |

**Regression Parameters**

| Parameter | Estimate | Std Error | 95% LCL | 95% UCL | t Statistic | P Level | Decision(0.05) |
|---|---|---|---|---|---|---|---|
| A | 0.88834 | 0.04855 | 0.78944 | 0.98724 | 18.296 | 0.00000 | Significant Parameter |
| C | -0.10223 | 0.03255 | -0.16854 | -0.03592 | -3.140 | 0.00362 | Significant Parameter |
| D | 45.63805 | 3.20963 | 39.10024 | 52.17585 | 14.219 | 0.00000 | Significant Parameter |

**Regression Summary**

| Iters | Log Likelihood | Psuedo R2 | Opt Threshold | F Statistic | Critical | P Level | Decision(0.01) |
|---|---|---|---|---|---|---|---|
| 11 | 54.91856 | 0.89998 | No | 1.07461 | 4.07403 | 0.38772 | Non-Significant Lack of Fit |

**Residual Analysis**

| Attribute | Method | Statistic | Critical | P Level | Decision(0.01) |
|---|---|---|---|---|---|
| Variances | Modified Levene | 6.92686 | 3.72540 | 0.00023 | Unequal Variances |
| Distribution | Shapiro-Wilk W | 0.93011 | 0.91004 | 0.03882 | Normal Distribution |

**Test Acceptability**

| Attribute | Statistic | Acceptable Range | Decision |
|---|---|---|---|
| Control Response | 0.96 | 0.9 - N/A | Passes acceptability criteria |

**Point Estimates**

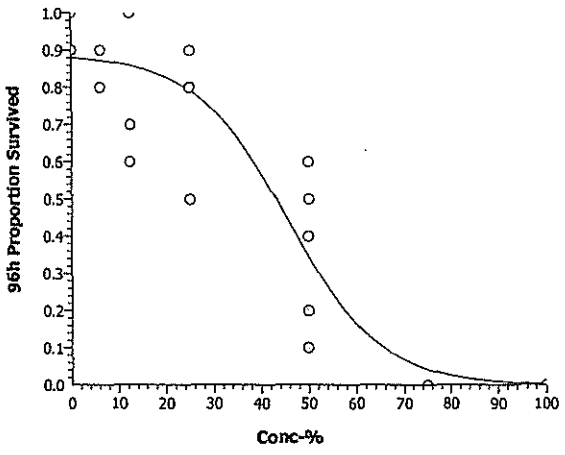
| % Effect | Conc-% | 95% LCL | 95% UCL |
|---|---|---|---|
| 50 | 44.05642 | 26.59244 | 58.86287 |

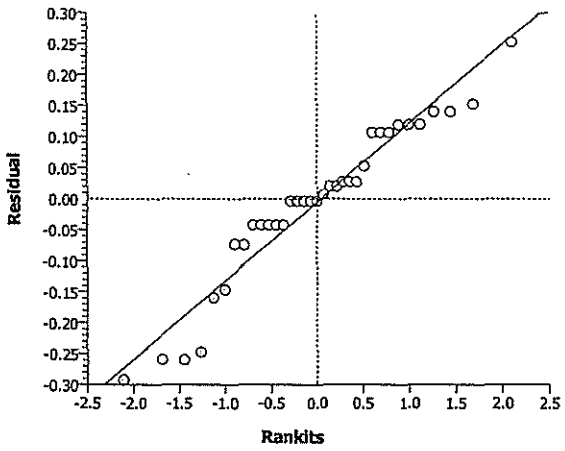
**Data Summary**

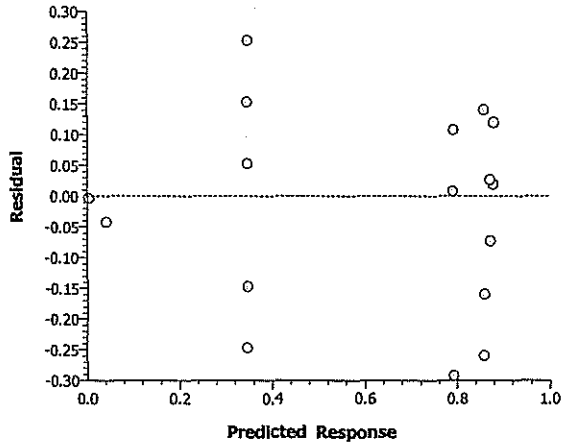
| | | | Calculated Variate(A/B) | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Conc-% | Control Type | Count | Mean | Minimum | Maximum | SE | SD | A | B |
| 0 | Lab Water | 5 | 0.96000 | 0.90000 | 1.00000 | 0.01118 | 0.05477 | 48 | 50 |
| 6.25 | | 5 | 0.86000 | 0.80000 | 0.90000 | 0.01118 | 0.05477 | 43 | 50 |
| 12.5 | | 5 | 0.78000 | 0.60000 | 1.00000 | 0.04183 | 0.20494 | 39 | 50 |
| 25 | | 5 | 0.80000 | 0.50000 | 0.90000 | 0.03536 | 0.17321 | 40 | 50 |
| 50 | | 5 | 0.36000 | 0.10000 | 0.60000 | 0.04233 | 0.20736 | 18 | 50 |
| 75 | | 5 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0 | 50 |
| 100 | | 5 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0 | 50 |

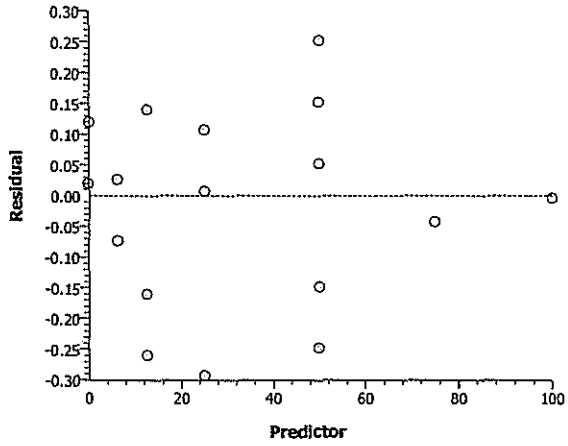
# CETIS Analysis Detail

Graphics



# Aquatic Research Organisms

## DATA SHEET

I. Organism History

Species: *Americamysis bahia*

Source: Lab reared ✓ Hatchery reared _____ Field collected _____

Hatch date 3-3-07 Receipt date _____

Lot number 030307MS Strain _____

Brood Origination *Florida*

II. Water Quality

Temperature 25 °C Salinity ~30 ppt DO —

pH 7.8 Hardness — ppm

III. Culture Conditions

System: *Recirc*

Diet: Flake Food ✓ Phytoplankton _____ Trout Chow ✓

Brine Shrimp ✓ Rotifers _____ Other *Encap. Shrimp Diet*

Prophylactic Treatments: _____

Comments: _____

IV. Shipping Information

Client: *ESI* # of Organisms: *280+*

Carrier: _____ Date Shipped: *3-6-07*

Biologist: *Mark Voorhees*

1 - 800 - 927 - 1650

PO Box 1271 • One Lafayette Road • Hampton, NH 03842 • (603) 926-1650

## EFFLUENT & DILUENT CHEMISTRY and WATER QUALITY DATA

| PARAMETER | 100% Effluent | 50% Effluent | Diluent - Lab Salt |
|---|---|---|---|
| TRC | <0.05 | | <0.05 |
| As Received - pH (SU) @ 20°C | 6.97 | | ~~25~~ 7.97 |
| As Received - Salinity (ppt) | 9.0 | | 25 |
| As Received - Dissolved Oxygen (mg/L)‡ | 0.7 | | 7.4 |
| As Received - Ammonia (pull) | -002 | | 15568-001 |
| Salinity Adjusted - pH (SU) @ 20°C | 7.44 | 7.62 | |
| Salinity Adjusted - Salinity (ppt) | 25 | 25 | |
| After Aeration - Dissolved Oxygen (mg/L) | 5.0 | 5.6 | |
| Salinity Adjusted - Ammonia (pull) | | -003 | |
| 48 hour Ammonia (pull) | -005 | ~~-005~~ -004 | 15568-002 |
| 48 hour pH (SU) @ 20°C | 8.27 | 8.07 | 7.59 |

‡ - Aerate prior to mixing concentrations.

## PREPARATION OF DILUTIONS

STUDY: 15566 | CLIENT: CH2M HILL - American Samoa

SPECIES: *A. bahia*

| Diluent: Lab Salt | Day: 0 Sample: EOA | | Day: 3 Sample: EOA | |
|---|---|---|---|---|
| Concentration | Vol. Eff. | Final Vol | Vol. Eff. | Final Vol |
| LAB | 0 | 1000 | 0 | 750 |
| 6.25% | 62.5 | | 46.87 | |
| 12.5% | 125 | | 93.75 | |
| 25% | 250 | | 187.5 | |
| 50% | 500 | | 375 | |
| 75% | 750 | | 750 | |
| 100% | 1000 | | | |

| HRS | Date | Time | Initials |
|---|---|---|---|
| 0 | 3/6/07 | 1530 | SJ |
| 48 | 3/8/07 | 1200 | CS |

Comments:

**AERATE SAMPLE PRIOR TO MIXING DILUTIONS AT START AND 48 HOURS.**

# RECORD OF METERS USED FOR WATER QUALITY MEASUREMENTS

| STUDY: 15566 | CLIENT: CH2M HILL - American Samoa |
|---|---|

WATER QUALITIES - A. bahia

| HOURS: | 0 | 24 | 48 - old | 48 - new | 72 | 96 |
|---|---|---|---|---|---|---|
| Water Quality Station # | 1 | 2 | 2 | 1 | 1 | 1 |
| Initials | SJ | CS | VT | CS | CS | CS |
| Date | 3/6/07 | 3/7/07 | 3/8/07 | 3/8/07 | 3/9/07 | 3/10/07 |

| Water Quality Station #1 | | Water Quality Station #2 | | COMMENTS |
|---|---|---|---|---|
| DO meter # | 3 | DO meter # | 19 | |
| DO probe # | 13 | DO probe # | 12 | |
| pH meter # | 1097 | pH meter # | 410 | |
| pH probe # | 49 | pH probe # | 48 | |
| S/C meter # | YSI30C | S/C meter # | 45B0C | |
| S/C probe # | 1 | S/C probe # | 1 | |
| Salinity meter # | | Salinity meter # | | |

Report No: 15566 SDG:
Project: CH2MHill - American Samoa

Sample ID: Effluent
Matrix: Water
Sampled: 03/08/07 0930

| Parameter | | Result | Quant Limit | Units | Date Prepared | Date of Analysis | Method/Reference |
|---|---|---|---|---|---|---|---|
| Ammonia-N | 15566-004 | 1.9 | 0.1 | mg/L as N | 03/13/07 | 03/13/07 | SM 4500-NH3 G |
| Ammonia-N | 15566-005 | 2.4 | 0.1 | mg/L as N | 03/13/07 | 03/13/07 | SM 4500-NH3 G |
| Ammonia-N | 15566-002 | 28 | 0.2 | mg/L as N | 03/09/07 | 03/09/07 | SM 4500-NH3 G |
| Ammonia-N | 15566-003 | 15 | 0.2 | mg/L as N | 03/09/07 | 03/09/07 | SM 4500-NH3 G |

Notes:

Report No: 15568 SDG:
Project: Diluent - Lab Salt 25 ppt

Sample ID: LAB SALT 25 ppt 03/0
Matrix: Water
Sampled: 03/08/07

| Parameter | | Result | Quant Limit | Units | Date Prepared | Date of Analysis | Method/Reference |
|---|---|---|---|---|---|---|---|
| Ammonia-N | 15568-002 | ND | 0.1 | mg/L as N | 03/13/07 | 03/13/07 | SM 4500-NH3 G |
| Ammonia-N | 15568-001 | ND | 0.1 | mg/L as N | 03/09/07 | 03/09/07 | SM 4500-NH3 G |

Notes:

ND = Not Detected



EnviroSystems, Inc. P.O. Box 778 Hampton, NH 03842-0778 603-926-3345 fax 603-926-3521 www.envirosystems.com

STUDY: 15566
CLIENT: CH2MHill - American Samoa
PROJECT: Wastewater Treatment Plant
TASK: Unionized Ammonia Calculations

| Day / Date | Treatment | Temperature Deg C | Sample pH SU | NH3 mg/L | Unionized NH3 mg/L |
|---|---|---|---|---|---|
| Day 0 | Lab Diluent | 20 | 7.97 | 0.10 | 0.004 |
| | 50% Effluent | 20 | 7.62 | 15.0 | 0.243 |
| | 100% Effluent | 20 | 6.97 | 28.0 | 0.103 |
| Day 2 | Lab Diluent | 20 | 7.59 | 0.1 | 0.002 |
| | 50% Effluent | 20 | 8.07 | 1.9 | 0.084 |
| | 100% Effluent | 20 | 8.27 | 2.4 | 0.165 |

ESI

EnviroSystems, Inc.
One Lafayette Road
P.O. Box 778
Hampton, NH 03843-0778
Telephone: 603-926-3345

# SAMPLE RECEIPT RECORD

ESI STUDY NUMBER: 15566 CLIENT: American Samoa

SAMPLE RECEIPT:
DATE: 3/6/07 TIME: 0930 BY: BB

DELIVERED VIA: ☐ FEDEX ☐ CLIENT ☐ ESI ☐ UPS ☐ OTHER

LOGGED INTO LAB:
DATE: 3/6/07 TIME: 1420 BY: SJ

SAMPLE CONDITION:

| | | |
|---|---|---|
| CHAIN OF CUSTODY: | ☒ YES | ☐ NO |
| CHAIN OF CUSTODY SIGNED: | ☒ YES | ☐ NO |
| CHAIN OF CUSTODY COMPLETE: | ☐ YES | ☒ NO |
| SAMPLE DATE: | ☒ YES | ☐ NO |
| SAMPLE TIME RECORDED: | ☐ YES | ☒ NO |
| SAMPLE TYPE IDENTIFIED: | ☒ YES | ☐ NO |
| CUSTODY SEAL IN PLACE: | ☒ YES | ☐ NO |
| SHIPPING CONTAINER INTACT: | ☒ YES | ☐ NO |
| SAMPLE TEMPERATURE (AT ARRIVAL): 12 °C | | |
| DOES CLIENT NEED NOTIFICATION OF TEMPERATURE? | ☐ YES | ☒ NO |
| SAMPLE ARRIVED ON ICE: | ☐ YES | ☒ NO |

COMMENTS: 1 5 gal Cubitainer

# CHAIN OF CUSTODY

| | | | | | | |
|---|---|---|---|---|---|---|
| PROJECT: | Joint Cannery Outfall Effluent Monitoring- JCO0701.NT | | | | | |
| FROM: | Karen Glatzel, gdc | | | | | |
| | P.O. Box 1238, Trinidad, CA, 95570-1238 | | | | 707-677-0123 | gdcocn@eathlink.net |
| TO: | Brian Buzby, EnviroSystems, Inc. | | | | | |
| | One Lafayette Road, Hampton, New Hampshire 03842 | | | | 603-926-3345 | |

| SAMPLE I.D. | DATE | TIME | MATRIX | NUMBER OF CONTAINERS | ANALYSIS REQUESTED: 96-hour acute mysid bioassay test | COMMENTS |
|---|---|---|---|---|---|---|
| JCO-07NT | 2/28/2007 | | Water | 1 | X | |
| | | | | | | |
| | | | | | | |
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| | | | |
|---|---|---|---|
| SAMPLED BY: K. Glatzel | | DATE/TIME: | 2/28/2007 |
| SHIPPED VIA: DHL | | DATE/TIME: | 3/1/2007 |
| RELINQUISHED BY: S.Costa | | DATE/TIME: | 3/1/2007 |
| RECEIVED BY: | | DATE/TIME: | 3/2/07 0930 |
| RELINQUISHED BY: | | DATE/TIME: | |
| RECEIVED BY: | | DATE/TIME: | |

SPECIAL INSTRUCTIONS/REMARKS:
Note - sample may be approximately 40% seawater
**Dilutions and aeration protocols as in previous test**

Rec'd at 12°C
Ice Melted